Nos. 16-2302, 16-2615

IN THE

United States Court of Appeals

FOR THE FEDERAL CIRCUIT

GILEAD SCIENCES, INC.,

Plaintiff-Cross-Appellant,

V.

MERCK & CO., INC., MERCK SHARP & DOHME CORP., AND
ISIS PHARMACEUTICALS, INC.,

Defendants-Appellants.

On Appeal from the United States District Court
for the Northern District of California, in No. 5:13-cv-04057-BLF

CORRECTED BRIEF FOR DEFENDANTS-APPELLANTS MERCK & CO., INC., MERCK SHARP & DOHME CORP., AND ISIS PHARMACEUTICALS, INC.

Bruce R. Genderson
Jessamyn S. Berniker
Stanley E. Fisher
Sanjiv P. Laud
Jessica Palmer Ryen
WILLIAMS & CONNOLLY LLP
725 Twelfth Street, N.W.
Washington, D.C. 20005
(202) 434-5000 (telephone)
(202) 434-5029 (facsimile)
bgenderson@wc.com

Jeffrey A. Lamken
Counsel of Record
Michael G. Pattillo, Jr.
Sarah J. Newman
James A. Barta
MOLOLAMKEN LLP
The Watergate, Suite 660
600 New Hampshire Avenue, N.W.
Washington, D.C. 20037
(202) 556-2000 (telephone)
(202) 556-2001 (facsimile)
jlamken@mololamken.com

Counsel for Merck & Co., Inc., Merck Sharp & Dohme Corp.,
and Isis Pharmaceuticals, Inc.

(Additional Counsel Listed on Inside Cover)

Stephen S. Rabinowitz
Mitchell Epner
HUGHES HUBBARD & REED LLP
One Battery Park Plaza
New York, NY 10004
(212) 837-6000 (telephone)
(212) 299-6807 (facsimile)
stephen.rabinowitz@hugheshubbard.com

Sara Margolis
MOLOLAMKEN LLP
430 Park Avenue, Floor 6
New York, NY 10022
(212) 607-8160 (telephone)
(212) 607-8161 (facsimile)

*Counsel for Merck & Co., Inc., Merck Sharp & Dohme Corp.,
and Isis Pharmaceuticals, Inc.*

UNITED STATES COURT OF APPEALS FOR THE FEDERAL CIRCUIT

Gilead Sciences, Inc. **v.** Merck & Co., Inc., et al.

Case No. 16-2302, 16-2615

CERTIFICATE OF INTEREST

Counsel for the:
☐ (petitioner) ☒ (appellant) ☐ (respondent) ☐ (appellee) ☐ (amicus) ☐ (name of party)

certifies the following (use "None" if applicable; use extra sheets if necessary):

1. Full Name of Party Represented by me	2. Name of Real Party in interest (Please only include any real party in interest NOT identified in Question 3) represented by me is:	3. Parent corporations and publicly held companies that own 10 % or more of stock in the party
Merck & Co., Inc.	None	None
Merck Sharp and Dohme Corp	None	Merck & Co., Inc.
Ionis Pharmaceuticals, Inc. f/k/a Isis Pharmaceuticals, Inc.	None	None

4. The names of all law firms and the partners or associates that appeared for the party or amicus now represented by me in the trial court or agency or are expected to appear in this court (**and who have not or will not enter an appearance in this case**) are:

Please see attached.

November 23, 2016
Date

/s/ Jeffrey A. Lamken
Signature of counsel

Please Note: All questions must be answered

Jeffrey A. Lamken
Printed name of counsel

cc: All counsel by ECF

Reset Fields

CERTIFICATE OF INTEREST

Appellants Merck & Co., Inc., Merck Sharp and Dohme Corp., and Ionis Pharmaceuticals, Inc. f/k/a Isis Pharmaceuticals, Inc., state that the following partners or associates have appeared on their behalf before the trial court or are expected to appear in this court:

From **Williams & Connolly LLP**: Bruce R. Genderson, Jessamyn Sheli Berniker, Stanley E. Fisher, Jessica Bodger Rydstrom, Sanjiv Prakash Laud, and Jessica Palmer Ryen.

From **Hughes Hubbard & Reed LLP**: Stephen Solomon Rabinowitz, James W. Dabney, Patrice Polyxene Jean, Wanda French-Brown, Mitchell E. Epner, David Lansky, and Stefanie Michelle Lopatkin.

From **Durie Tangri LLP**: Joshua H. Lerner and Laura Elizabeth Miller.

From **Fried, Frank, Harris, Striver and Jacobson LLP**: Randy C. Eisensmith and Naz Erdeniz Wehrli.

From **Sidley Austin LLP**: Todd L. Krause, S. Isaac Olson, David T. Pritikin, M. Patricia Thayer, and Sue Wang.

From **MoloLamken LLP**: Jeffrey A. Lamken, Michael G. Pattillo, James A. Barta, Sara E. Margolis, and Sarah J. Newman.

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Black's Law Dictionary (3d ed. 1933) 44

Dobbs, *Law of Remedies* (2d ed. 1993) 38, 45, 46

Restatement (Second) of Torts (1979) 38, 39, 41

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STATEMENT OF RELATED CASES

Pursuant to Federal Circuit Rule 47.5, Defendants-Appellants Merck & Co., Inc., Merck Sharp & Dohme Corp., and Isis Pharmaceuticals, Inc. note that:

(a) there have been no other appeals in this case; and

(b) aside from the present case, there are no other cases pending in this or any other court that will directly affect or be directly affected by this Court's decision in No. 16-2302 or No. 16-2615.

JURISDICTIONAL STATEMENT

The district court had jurisdiction under 28 U.S.C. §§1331 and 1338(a). Final judgment was entered on August 16, 2016. Merck & Co., Inc., Merck Sharp & Dohme Corp., and Isis (now Ionis) Pharmaceuticals, Inc. (collectively "Merck") timely appealed on August 23, 2016, and Gilead Sciences, Inc., cross-appealed on September 2, 2016. The cases were consolidated. This Court has jurisdiction under 28 U.S.C. §1295.

STATEMENT OF THE ISSUES

The district court entered summary judgment on infringement after Gilead conceded that use of its accused products satisfied every limitation of the asserted claims in Merck's patents, U.S. Patent No. 7,105,499 ('499 patent) and U.S. Patent No. 8,481,712 ('712 patent). After a trial, the jury found the patents not invalid and awarded Merck $200 million in damages. The questions presented are:

1. Whether the district court properly nullified the jury's verdict by barring Merck from asserting the '499 patent against Gilead under the doctrine of unclean hands.

2. Whether the district court properly nullified the jury's verdict by barring Merck from asserting the '712 patent against Gilead for unclean hands to ensure Merck suffered a "penalty."

PRELIMINARY STATEMENT

The decision below invoked the exceptional, and rarely applied, defense of unclean hands to overturn a $200 million jury verdict. The court invoked that doctrine for the ’712 patent without finding business or litigation misconduct relating to that patent in any way. The court admitted, “it doesn’t appear that there’s any evidence” regarding “the ’712.” But the court insisted the ’712 patent must be unenforceable to ensure Merck suffered a “penalty.”

With respect to the ’499 patent, the court faulted a Merck patent prosecutor, Philippe Durette, for participating in a 2004 due-diligence call where he was exposed to a competitor’s confidential compound. But the ’499 patent’s claims already covered that compound ***before*** the call occurred. And Durette did nothing with respect to the ’499 patent until all confidentiality restrictions expired. Ultimately, the court was incensed by what it deemed untruthful testimony by Durette—long-since a former employee—about whether he participated in the 2004 due-diligence call. But that testimony was irrelevant, and there is no evidence Merck ever encouraged or controlled it. To the contrary, Merck agreed Durette was on the call. Far from seeking to exploit the testimony, Merck sought its exclusion; Gilead insisted on putting it before the jury.

The district court’s decision to impose unclean hands defies centuries of law. The unclean-hands doctrine ensures courts do not abet iniquity by helping parties reap the benefits of their misconduct. It is not a license to punish conduct that neither confers an unfair advantage on one party nor unfairly disadvantages the other. The court below, however, refused even to require that the conduct be "material," calling any materiality requirement "non-existent." The alleged misconduct, moreover, did not reflect a deliberately planned and carefully executed scheme by Merck to defraud or deceive. In denying enforcement, the court below relied on clearly erroneous factual assertions. And its ruling denies rather than pursues equity. It unjustly enriches an infringer by hundreds of millions of dollars and licenses it to continue infringing with impunity.

STATEMENT OF THE CASE

I. FACTUAL BACKGROUND

A. Merck Discovers a Revolutionary Class of Compounds for Treating Hepatitis C

Merck's '499 and '712 patents concern treatment of Hepatitis C. Approximately 200 million people are infected with the Hepatitis C virus ("HCV"). Appx00152 (1:29-38). For years, treatments were ineffective and produced severe side effects. Appx19900-19901 (199:6-201:1). Merck is a leading innovator in anti-virals, including HIV drugs. Appx20289-20290 (941:12-945:23). In 1998, Merck partnered with Isis Pharmaceuticals to revolutionize HCV treatment. Appx20291 (948:19-950:21).

HCV reproduces by hijacking the host liver cell's machinery. Appx06774. Using a viral enzyme (a "polymerase"), HCV takes naturally occurring nucleosides—the "building blocks" of genetic code—and assembles strands of its own RNA. Appx06772; Appx06774. Nucleosides consist of a "sugar" bonded to a "base." Appx06765; Appx07662. Scientists at Merck recognized that, by altering the structure of a naturally occurring nucleoside, it was possible to make nucleoside "inhibitors" or "chain terminators" that stop viral replication mid-stream. Appx20291-20293(951:15-956:25). A chain terminator has a chemical structure sufficiently similar to a natural nucleoside that the viral polymerase will incorporate it into the RNA chain. But it is sufficiently different that it prevents other nucleosides from being added to complete that strand. Appx19912(247:20-248:6).

Merck's Dr. David Olsen led the collaboration with Isis to find chain terminators for HCV. Appx20291-20292(949:25-952:17); Appx20294(962:16-17). Over several years, Merck tested more than 2,000 nucleoside analogs, including 1,000 novel compounds Isis made. Appx20296(970:21-971:2). Merck discovered many potent inhibitors of HCV replication. Appx20305-20306(1005:2-1009:10). It found inhibitors active at concentrations of less than 100 micromolar, more than 50 of which were active at less than 10 micromolar. Appx20305(1005:17-23). That was "a significant breakthrough, eureka moment." Appx20479(1316:13-21). With the benefit of the structure-activity relationships discovered through this testing, Merck identified a class of compounds believed to have anti-HCV activity. Appx20297 (972:2-21).

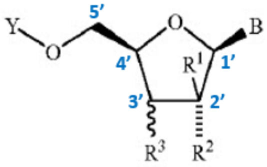
B. The Applications Leading to the ’499 Patent

Recognizing Merck had “discovered an important class of molecules,” Dr. Olsen “felt it was important to . . . protect that invention.” Appx20297 (973:15-23). Philippe Durette—a Merck chemist turned patent prosecutor—was assigned to handle patent prosecution. Appx19933 (327:24-328:9); Appx20301 (991:2-16).

On January 22, 2001, Merck filed a provisional patent application disclosing and claiming a class of nucleoside analogs with desirable biological activity and their use to treat HCV. Appx25804; Appx20301 (988:4-989:12). On January 18, 2002, Merck filed two PCT applications and a non-provisional application, all claiming priority to the provisional application. Appx24832; Appx26913; Appx00223. One PCT application, which published on July 25, 2002, Appx24832, issued as the ’499 patent, Appx00150. The non-provisional application led to the ’712 patent. Appx00223.

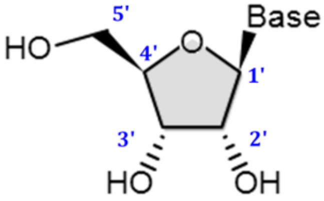
1. *The Specification*

The shared specification of the ’499 and ’712 patents explains that the compounds of the invention are useful as inhibitors of HCV polymerase and HCV replication, and for treatment of HCV. Appx00152 (1:21-25). The specification defines classes of compounds using generic chemical formulas, such as:



Appx00158 (13:7-14) (annotations in blue). The blue numbers above correspond to positions on the molecule—1' through 5'—where different atoms might be attached. To account for the molecule's three-dimensional structure, the specification designates "R^1" as "up" and "R^2" as "down" in the 2' position; while "R^3" could be up or down. "B" denotes the base.

The four naturally occurring RNA nucleosides have hydroxyl (OH) groups at the 2' and 3' down positions:



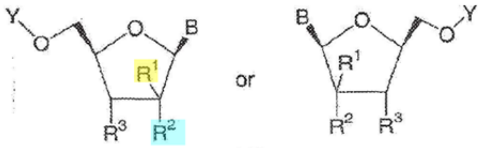
Appx32781 (annotations in blue). Two of those nucleosides have "single-ring" bases, and two have "double-ring" bases. Appx06767-06768; Appx20633 (1552:3-7).

The specification includes 149 Examples with modifications to the naturally occurring nucleosides, particularly at the 2' and 3' positions. Appx00171-00217; Appx20634-20635 (1556:18-1559:25). The specification reports that testing of the representative compounds revealed anti-HCV activity of less than 100 micromolar, Appx00217-00218 (132:56-57, 133:22-23), which was "quite impressive" in 2002, Appx20479 (1316:13-25).

2. *The Applications' Inclusion of 2' Methyl-Up/Fluoro-Down Configurations*

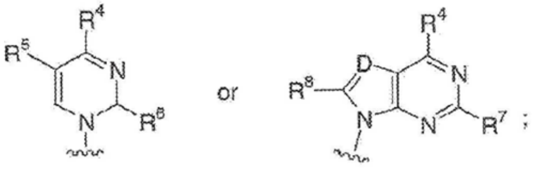
The active form of the infringing compounds in this case are nucleosides having a single-ring base and a sugar with methyl up and fluoro down in the 2' position. There is no dispute that Merck filed patent applications in 2001 and 2002, including the PCT that issued as the '499 patent, that claimed their use. *See* Appx20214 (824:7-22); Appx20218 (841:3-21).

The applications claimed classes of compounds using Markush groups. Claim 6 of the January 2001 provisional, for example, covers a genus that encompasses a compound with a 2' methyl-up/fluoro-down sugar with a single- or double-ring base:



(III)

wherein B is



D is N, CH, C-CN, C-C_{1-3} alkyl, C-CONR^{11}R^{11};

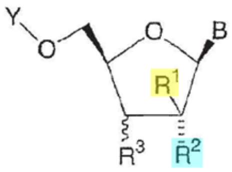
Y is H or P(O)R^{9}R^{10};

R^1 is hydrogen or methyl and one of R^2 and R^3 is OH and the other of R^2 and R^3 is selected from the group consisting of

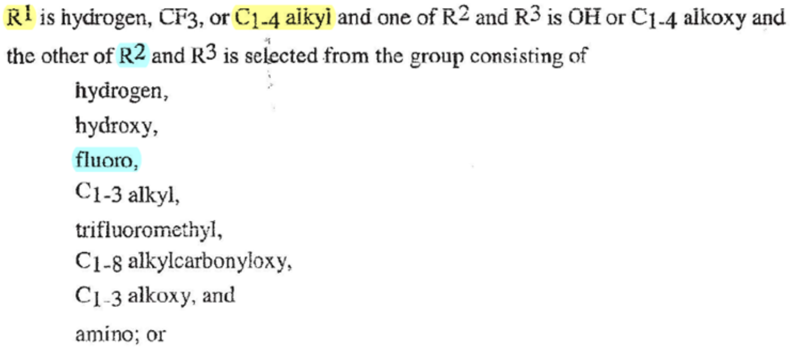
hydrogen,
hydroxy,
fluoro,
C_{1-3} alkyl,
C_{1-3} alkoxy, and
amino; or

Appx25954-25955 (claim 6, excerpt, colors added). So, in this example, R^1 (2' up) encompasses methyl, while R^2 (2' down) could be fluoro.

Claim 6 of the 2002 PCT application is similar (referring to methyl as "C_1 alkyl"):



(III)



R^1 is hydrogen, CF_3, or C_{1-4} alkyl and one of R^2 and R^3 is OH or C_{1-4} alkoxy and the other of R^2 and R^3 is selected from the group consisting of

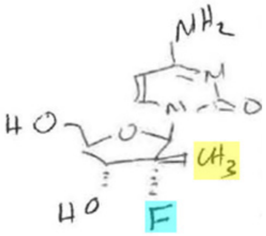
hydrogen,
hydroxy,
fluoro,
C_{1-3} alkyl,
trifluoromethyl,
C_{1-8} alkylcarbonyloxy,
C_{1-3} alkoxy, and
amino; or

Appx25036-25037 (claim 6, excerpts, colors added); Appx20633 (1551:6-10). PCT Claim 8 depends from Claim 6 and is limited to single-ring bases. Appx25038.

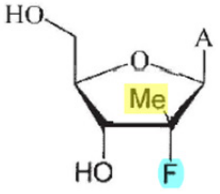
Dr. Olsen "made sure" Merck's first provisional application covered nucleoside analogs with that 2' methyl/fluoro configuration. Appx20297 (973:1-975:16). By May 2000, Merck had made the "important finding" that 2' fluoro compounds were "substrates" the HCV polymerase would incorporate into the RNA chain during replication. Appx20299 (980:10-982:2); Appx30348-30349. And by the fall of 2000, Merck made the critical finding that 2' methyl-up compounds inhibit

HCV replication. Dr. Olsen identified those compounds as a "top priority." Appx20296-20297(971:3-973:23); Appx20285(926:6-16); Appx30371.

By 2001, Dr. Olsen's team had depicted and discussed nucleoside analogs "combin[ing]" "a methyl up at 2' and a fluoro down at 2'." Appx20285(924:9-925:12); Appx20299(981:25-982:8). In early 2001, another named inventor, Isis's Dr. Thazha Prakash, sketched a 2' methyl-up ("CH_3"), 2' fluoro-down compound with a single-ring base in his lab notebook:

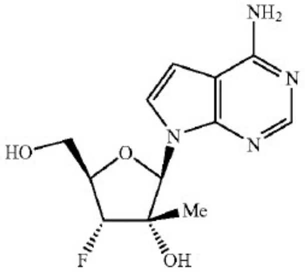


Appx32431 (colors added); Appx20211(812:1-814:14). Isis chemist Dr. Quanlai Song presented a 2' methyl-up/fluoro-down compound with a double-ring base at an April 2001 meeting:



Appx22824; Appx22879 (colors added). He made the compound in an intermediate, "protected" form. Appx32315(149:19-151:17); Appx32285(121:06-122:23).

Although these compounds were claimed in the Markush groups in the applications, testing was limited to structurally similar compounds, including a 2' methyl-up/3’ fluoro-down compound, which was active and included as Example 143 in the specification:



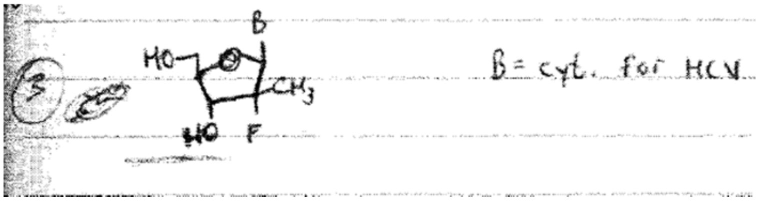
Appx00213 (123:5-15); Appx20311 (1029:14-1030:2).

C. Gilead’s Predecessor, Pharmasset, Fails To Develop an Active Compound Until It Reviews Merck’s Applications

A company called Pharmasset had been working on anti-HCV compounds since 1999. It had not made a single novel active compound by the time Merck’s PCT applications published in July 2002. Appx20036 (488:5-7); Appx20050-20051 (544:21-545:11).

Four days after Merck’s applications published, Pharmasset’s Executive Director, Dr. Schinazi, asked in-house lawyer Bryce Roberts to obtain them for review “ASAP.” Appx31083; Appx20046 (525:16-527:17). Roberts forwarded the applications to Dr. Schinazi and Pharmasset employee Alan Roemer. Appx31083. It was “standard operating procedure” at Pharmasset to monitor patent literature. Appx20047-20048 (531:20-533:7). But Pharmasset was not merely following “the Hepatitis C landscape.” Appx19963 (448:17-19). According to Dr. Michael Otto, Pharmasset's Chief Scientific Officer, Pharmasset was "trying to cash on the loopholes" in other companies' patents. Appx24823; Appx20037 (490:4-5).

In November 2002, Dr. Otto challenged Pharmasset chemists to "find loopholes" in Merck's applications. Appx20048 (533:11-19). Days later, chemist Jeremy Clark came to Dr. Otto's office. Holding a copy of Merck's application, Clark told Dr. Otto of his idea for a compound now known as PSI-6130. Appx20040 (502:8-503:9); Appx20048 (533:20-534:4). The compound Clark drew in November 2002 (shown below from Clark's lab notebook) is the same compound Isis's Dr. Prakash had drawn in early 2001:



Appx24619; Appx32292-32293 (32:11-37:12); *compare* p. 9, *supra*. It has the same 2' methyl-up/fluoro-down sugar as the compound Dr. Song identified and made in 2001. *See* p. 9, *supra*.

In December 2002, Pharmasset sought a National Institutes of Health grant, citing one of Merck's 2002 patent applications as providing Pharmasset's "rationale" for pursuing 2' methyl-up compounds. Merck's application, it explained, taught that such compounds "showed a potent anti-HCV activity." Appx31499 (citing n.35); Appx31507 n.35 (citing WO 02/057287); Appx31084.

Clark synthesized and tested PSI-6130 by May 2003. Appx20040(504:8-11); Appx20041(508:15-21). "[PSI-]6130 [was] the first compound" Pharmasset made "that is active against HCV." Appx20050-20051(544:21-545:8).

D. Pharmasset's Effort To License PSI-6130 to Merck Fails Because Merck/Isis Already Had That Compound "In Their Stable"

PSI-6130 did not fall within any "loopholes" in Merck's application. Appx24823. In May 2003, Dr. Otto confirmed to Roemer and Dr. Schinazi that "there is methyl up and fluorine down in both the 2' and 3' positions" in Merck's application. Appx31458; Appx20051(545:12-546:12). Dr. Otto "knew" that "[y]ou could find [PSI-6130] in the claims, absolutely." Appx20050(543:13-20). Nonetheless, in early 2004, Pharmasset approached Merck about partnering to develop PSI-6130 as a "clinical candidate." Appx32369; *see* Appx20500(1402:5-24). Pharmasset was "seeking . . . 'in excess of $100 million.'" Appx23706.

1. *The Non-Disclosure Agreements*

The companies executed non-disclosure agreements. *See, e.g.*, Appx32151-32155. Merck agreed not to disclose Pharmasset's "[c]onfidential [i]nformation" to third parties, Appx32153(¶5), or to use confidential information except to evaluate the potential collaboration, Appx32153(¶6). The NDA, however, included standard carve-outs. It provided that "Confidential Information shall not . . . include information which . . . is in the public domain" or lawfully "becomes part of the public domain." Appx32152(¶3(ii)).

2. *The March 17, 2004 Due-Diligence Call*

After receiving a Pharmasset term sheet, Merck's Business Development Executive Director, Pamela Demain, emailed a responsive term sheet to Merck's team. Appx23706. Because "Pharmasset ha[d] not yet permitted [Merck] to review the structure of PSI 6130," *id.*, Merck's term sheet was "based on [several] assumptions," one being that PSI-6130 "is a chain terminator of HCV polymerase," Appx32370. Demain stated that "Phil Durette will view the structure during a patent due diligence meeting." Appx23706.

Durette and Douglas Pon conducted the due-diligence call with Pharmasset's Dr. Schinazi, Roberts, and Roemer on March 17, 2004. Appx31544. According to Roemer's "real-time contemporaneous" notes, Appx19958(429:16-21), Merck's "objective[s]" included determining whether Pharmasset's compound was "[n]ovel," and whether it was covered by non-Pharmasset patents that might limit "[f]reedom to operate." Appx31544. Durette asked Pharmasset whether its compound was covered by "3rd party patents." Appx31544-31545. Pharmasset responded, "Yes, which we've licensed." Appx31544. Dr. Schinazi referenced "broad claims" in his own 1999 patent. *Id.* Durette sought to confirm that "[o]ther than the broad claim, there are no other 3rd party" patents, and someone specified "Roche, ***Merck***, Idenix, ***Isis***" as possibilities. Appx31545 (emphasis added). Although Dr. Schinazi knew Merck's patent application covered Pharmasset's compound, *see* p. 12, *supra*, he said nothing.

The parties confirmed that a "[f]irewall" covered the conversation. Appx31544. When Pharmasset disclosed a 2' methyl-up compound, Appx31545, Durette interjected that "[i]t's a problem"—he needed to speak with his supervisor because it "'seems quite related to things that I'm involved with.'" *Id.* Pharmasset nonetheless "described" PSI-6130. *Id.* Durette clarified he was "personally conflicted; not the company." *Id.* Toward the end, Roemer's notes state: "Full disclosure of the compound structure to Phil Durette and Doug Pon, both of whom were asked if they were firewalled; they both said that they were within the firewall." *Id.* After the call, Durette removed himself from the due diligence. Appx19944(372:24-373:15).

3. *Negotiations Fail Because Merck's Patent Already Covered PSI-6130*

A week later, Merck and Pharmasset held another call. Merck had since identified PSI-6130's "[o]verlap within the genus" claimed in Merck's patent. Appx32188. Merck warned Pharmasset that it did not have "freedom to operate." *Id.* Pon explained that, "when [Merck] started the negotiations, we assume[d] that [PSI-6130] was novel." *Id.* The negotiations failed. As Dr. Schinazi acknowledged, "Merck stepped down" because they "weren't very happy" that Pharmasset was attempting to "licens[e] a compound that [Merck] already had in their stable."

Appx20341-20342(1151:11-1152:1). Pharmasset later licensed PSI-6130 to Roche, and Roche obtained a license from Merck (with Pharmasset's consent). Appx20504(1415:23-1418:15); Appx32590.

E. Pharmasset's "Clark" Application Covering PSI-6130 Publishes in 2005

On April 21, 2004, Pharmasset filed the "Clark" PCT application, claiming compounds that included PSI-6130; it published on January 13, 2005. Appx26998. All the final compounds described in Clark, and all those recited in its 130 claims, were 2' methyl-up/fluoro-down nucleosides. Appx27093-27221. The first four Examples were synthetic methods for making PSI-6130 and other 2' methyl/ fluoro nucleosides. Appx27074-27085.

PSI-6130 was the first Example; it was also the only compound for which biological data was reported (with two controls). Appx27074-27075; Appx27085-27092 (Example 5). The data showed PSI-6130 was highly potent at ~2 micro-molar:

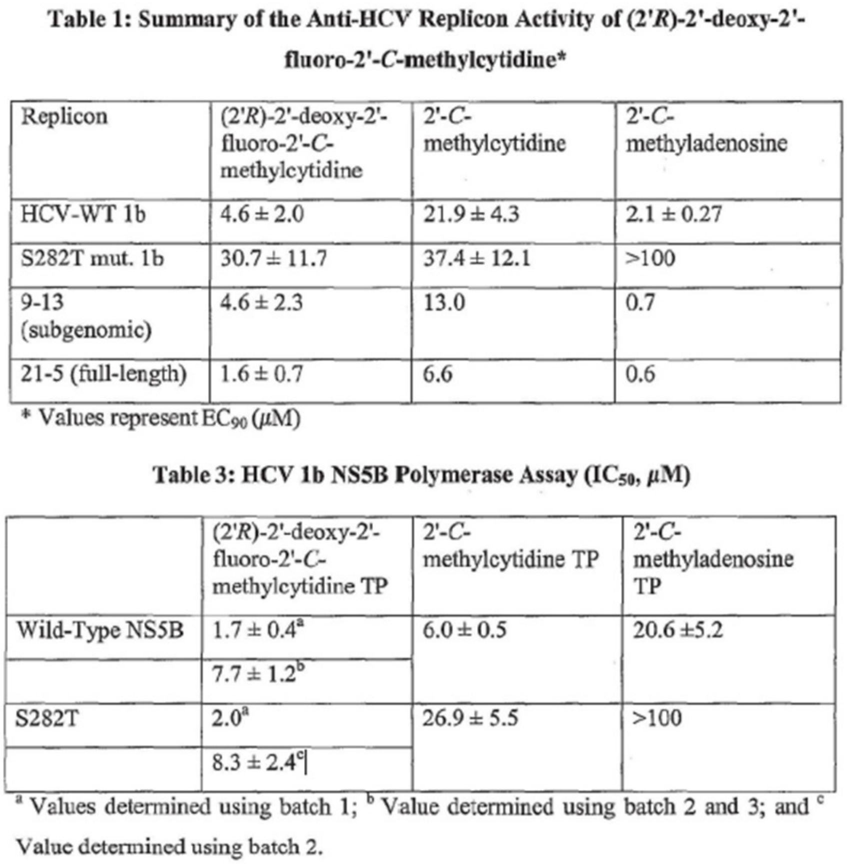


Table 1: Summary of the Anti-HCV Replicon Activity of (2'*R*)-2'-deoxy-2'-fluoro-2'-*C*-methylcytidine*

Replicon	(2'*R*)-2'-deoxy-2'-fluoro-2'-*C*-methylcytidine	2'-*C*-methylcytidine	2'-*C*-methyladenosine
HCV-WT 1b	4.6 ± 2.0	21.9 ± 4.3	2.1 ± 0.27
S282T mut. 1b	30.7 ± 11.7	37.4 ± 12.1	>100
9-13 (subgenomic)	4.6 ± 2.3	13.0	0.7
21-5 (full-length)	1.6 ± 0.7	6.6	0.6

* Values represent EC_{90} (μM)

Table 3: HCV 1b NS5B Polymerase Assay (IC_{50}, μM)

	(2'*R*)-2'-deoxy-2'-fluoro-2'-*C*-methylcytidine TP	2'-*C*-methylcytidine TP	2'-*C*-methyladenosine TP
Wild-Type NS5B	1.7 ± 0.4^a	6.0 ± 0.5	20.6 ±5.2
	7.7 ± 1.2^b		
S282T	2.0^a	26.9 ± 5.5	>100
	8.3 ± 2.4^c		

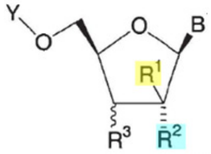
[a] Values determined using batch 1; [b] Value determined using batch 2 and 3; and [c] Value determined using batch 2.

Appx27088-27089 (Tables 1 and 3); Appx19948-19949 (389:23-390:14).

F. The Confidentiality Obligation Ends and Durette Narrows the ’499 Patent’s Claims

Consistent with industry practice, Merck and Durette monitored patent applications in their areas. Appx19949 (390:23-391:9); Appx20306 (1010:12-17). Durette testified that, when Clark publicly disclosed the structure and activity of PSI-6130 in 2005, he would have understood that “obligations under the [Pharmasset] confidentiality agreement terminated immediately.” Appx19949 (390:18-19); *see* p. 12, *supra*.

Between the March 2004 due-diligence call and Clark's publication, no substantive action occurred on Merck's '499 application. Shortly after Clark published, Durette amended Merck's '499 application to narrow the claims at the 2' down and 3' positions, focusing more specifically on compounds with 2' or 3' fluoro, a class that included PSI-6130. Appx28319-28320. The prior claims had fluoro as one of several potential options at the 2' down or 3' positions:

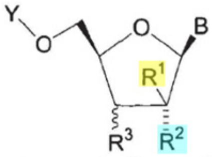


R^1 is hydrogen, CF_3, or $C_{1\text{-}4}$ alkyl and one of R^2 and R^3 is OH or $C_{1\text{-}4}$ alkoxy and the other of R^2 and R^3 is selected from the group consisting of

hydrogen,
hydroxy,
fluoro,
$C_{1\text{-}3}$ alkyl,
trifluoromethyl,
$C_{1\text{-}8}$ alkylcarbonyloxy,
$C_{1\text{-}3}$ alkoxy, and
amino; or

Appx27482-27483 (claim 44, excerpts, colors added).

Cancelling that claim, Durette submitted an otherwise identical claim that removed options at the 2' down and 3' positions to require fluoro at one of those positions:



R^1 is hydrogen, CF_3, or $C_{1\text{-}4}$ alkyl and one of R^2 and R^3 is OH or $C_{1\text{-}4}$ alkoxy and the other of R^2 and R^3 is fluoro;

Appx28319-28320 (claim 53, excerpts, colors added). After additional amendments not relevant here, the ’499 patent issued on September 12, 2006. Appx00150.

G. Merck’s ’712 Patent and Sofosbuvir

On February 2, 2007, Durette filed the application that ultimately issued as the ’712 patent. Appx24147. Its ancestor was a non-provisional application filed on January 18, 2002 (not the PCT that issued as the ’499 patent). *See* Appx00223. Unlike the ’499, the claims of the ’712 application, as filed in 2007, did ***not*** cover PSI-6130. Appx24150-24152. Durette had no further involvement in the ’712 patent’s prosecution; he retired from Merck in June 2010. Appx19954(413:18-21).

In 2010, Pharmasset published an article describing a new compound for treating HCV, a prodrug now known as “sofosbuvir.” Appx31990-32007. In the liver, sofosbuvir metabolizes into an active 2’ methyl-up/fluoro-down compound. Appx19913(249:18-252:11).

In 2011, Jeffrey Bergman took over prosecuting Merck’s ’712 application. Appx32383(17:25-20:08). Bergman was not involved in the 2004 Merck-Pharmasset due-diligence call. In July 2011, Bergman amended the ’712 application, adding claims that covered sofosbuvir metabolites Pharmasset published in 2010.

Appx24394-24410; Appx32386-32387 (56:19-59:01). The ’712 patent issued on July 9, 2013. Appx00223.

II. PROCEEDINGS BELOW

After acquiring Pharmasset, Gilead obtained FDA approval of Sovaldi® for treating HCV. Appx19903 (212:4-20). Sovaldi®’s active ingredient is sofosbuvir. Later, Gilead obtained FDA approval for Harvoni®, a combination of sofosbuvir and another HCV inhibitor. Appx19906 (223:7-25); Appx30120.

On August 5, 2013, before Sovaldi® came to market, Merck offered to discuss licensing the ’499 and ’712 patents to Gilead “for commercialization of sofosbuvir.” Appx32412. On August 30, Gilead filed a complaint seeking declaratory judgments of non-infringement and invalidity of both patents. Appx00136. Merck counterclaimed for infringement. Appx00460-00463 (¶¶ 11-34); Appx00947 n.1; Appx00951 (¶¶ 35-43).

A. Pretrial Proceedings

1. *Gilead Effectively Concedes Infringement but Presses Invalidity and Unenforceability*

Gilead eventually stipulated that, under the district court’s claim construction, using Sovaldi® and Harvoni® in accordance with their labels resulted in use of the methods and compounds recited in the asserted claims of Merck’s ’499 and ’712 patents. Appx01899; Appx07514 & n.1. The court granted Merck summary judgment of infringement. Appx15677.

Gilead pursued invalidity defenses, including lack of enablement and lack of written description. Appx15747-15774. Gilead also claimed that Merck "derived the invention from Jeremy Clark" through the failed business discussions and from the Clark application published in 2005. Appx15740. Gilead asserted unclean hands based on the same theory—that Merck "obtain[ed] its patent rights by deriving the invention from Pharmasset's confidential disclosures." Appx15735.

While disputing those contentions, Appx08106-08135, Merck urged that Gilead's written-description and enablement challenges rendered the derivation defense irrelevant, Appx15804-15810. If the January 18, 2002 specification adequately described and enabled the asserted claims—affording Merck priority to the invention as of 2002—then Merck had conception as of then and could not possibly have derived its invention from Pharmasset during a meeting years later in 2004. Appx15804-15810.[1] Conversely, if the January 2002 specification failed to describe and enable the claims, they would be invalid, and the derivation defense would be moot. Appx15809. Gilead's unclean-hands defense likewise rested on the theory that Merck derived the compounds described in its 2002 application through events in 2004-2005. *See* Appx15735.

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[1] Filing a patent application establishes conception of the subject matter in the application as of that date. *Hyatt v. Boone*, 146 F.3d 1348, 1352 (Fed. Cir. 1998). If the January 2002 specification satisfied "the written description and enablement requirements," the asserted claims were entitled to January 18, 2002 as the date of "conception." *Frazer v. Schlegel*, 498 F.3d 1283, 1287 (Fed. Cir. 2007).

2. *Gilead Deposes Durette and Cancels the Deposition of Merck's Designated 30(b)(6) Witness on the Due-Diligence Call*

In response to interrogatories asking Merck to "detail [its] communications with Pharmasset . . . including with respect to PSI-6130," Appx22266, Merck referred Gilead to Merck documents showing that "the March 17, 2004 telephone conference regarding PSI-6130" included "Phil Durette," Appx22212 (¶8). Gilead notified Merck that it intended to depose Durette (even though Merck had not identified Durette as a potential trial witness). Appx22211 (¶¶2-4). In response to Gilead's Rule 30(b)(6) deposition notice for testimony on 43 topics, Merck designated Durette as its representative on prosecution of the '499 patent and 13 other patents. Appx22214-22216 (¶¶13-21). Merck did not designate Durette as its 30(b)(6) corporate witness regarding the '712 patent prosecution or any aspect of the PSI-6130 due diligence, including the March 2004 due-diligence call. It designated Stephen de Laszlo on that due diligence. Appx22394.

Gilead's questioning of Durette at his deposition focused extensively on the March 2004 due-diligence call from 11 years earlier. Durette's responses were at best ambiguous. Early on, Durette stated only that he did not "recall such a conversation." Appx22336 (19:1-3). Gilead's counsel repeatedly circled back, and Durette eventually asserted he was not on the call. He was "positive" he "never saw a structure of the Pharmasset compounds until it published later." Appx22339 (30:21-31:10). When Gilead showed Durette an email stating that Durette would participate in a due-diligence meeting, Appx22340(33:13-24 (discussing Appx23706-23707)), Durette responded that his "memory of this involvement is not very strong," Appx22340(35:4-5); *see* Appx22340(35:14-16). He testified that "[e]vidently I reviewed a term sheet," Appx22340(36:18), but maintained he "never participated in a due diligence meeting," Appx22341(37:13-18).

Durette eventually returned to his original answer—that he did not "recall being assigned to do patent due diligence by Merck." Appx22374(172:15-16). Eleven times he repeated that he did not "recall participating in that phone conversation." Appx22374(172:22-23); *see* Appx22374-22375(172:24-174:21). At the end of the deposition, Gilead's counsel stated, "I know you don't remember it. What I'm asking you is: . . . [C]ould [the call] have happened and you just don't remember?" Appx22380(194:9-11). Durette responded: "I guess if I don't remember, both possibilities are possible." Appx22380(194:14-15).

Durette explained that Merck had a policy against due diligence being performed by attorneys prosecuting a "docket in a related area," because "potential issues" could create an "appearance of impropriety" or "conflict." Appx22341 (38:1-13, 39:3-40:4). But published, non-confidential information in the public domain was still free for all to use. Appx22380(195:21-24).

Durette testified that he saw the Clark application around the time he amended the '499 patent's claims in February 2005, but did not recall precisely when. Appx22343-22344 (48:10-52:1); Appx22348 (66:24-68:24). He testified that he was waiting for PTO action when he made the amendments narrowing the claims, and made them "[t]o expedite the prosecution," because "limiting the scope" of the claims might raise fewer potential "issue[s] of patentability." Appx22347 (62:14-21).

Gilead chose not to depose any other Merck witness about the due-diligence call. Following Durette's deposition, Gilead cancelled the deposition of Merck's corporate representative regarding the call, stating it "has elected to forgo the 30(b)(6) deposition of Mr. De Laszlo on [those] topics." Appx22396. Gilead had previously cancelled the deposition of Pon, the other Merck participant on the call. Appx22213-22214 (¶¶ 11-12).

3. *Merck Unsuccessfully Seeks To Exclude Durette's Testimony and Post-January 18, 2002 Evidence as Irrelevant and Prejudicial*

Merck did not adopt Durette's testimony about the due-diligence call. It filed motions *in limine* to exclude evidence regarding events after January 18, 2002—including Durette's testimony about the March 2004 due-diligence call and the 2005 amendment of the '499 patent—as legally irrelevant. *See* Appx15720-15726; Appx15804-15810. If the priority date for its patents was January 18, 2002, Merck explained, it could not have "derived" its invention from the March 2004 call. And if Merck was not entitled to that priority date, then the claims were invalid in any event. *See* p. 20 & n.1, *supra*. Merck also urged that the due-diligence-call evidence was "unfairly prejudicial" to Merck. Appx15722. The court agreed that Durette's testimony "certainly puts Merck in a bad light." Appx19413 (79:10-80:1). But it denied Merck's motion because the call was "relevant to Gilead's derivation defense." Appx19220-19222.

B. The Jury Rejects Gilead's Invalidity Defenses and Awards $200 Million in Damages

Gilead's case. Gilead's case put Durette front and center. "You're going to hear from Mr. Durette," Gilead told the jury in opening, "because we're calling him in our case." Appx19885 (137:5-6). Gilead said it would prove "Durette [was] on the phone with Pharmasset" in March 2004, "learn[ed] the structure of PSI-6130," Appx19886 (143:2-4), and used that "inside information" to write claims covering Pharmasset's work, Appx19886-19887 (143:24-146:19).

Gilead questioned Durette extensively in its case-in-chief. *See* Appx19583. It purported to impeach Durette numerous times with prepared video clips from his deposition. *See* Appx19937-19939 (345:8-347:7, 349:7-350:4, 352:4-12; 353:11-21); Appx19942 (362:15-24). Durette testified that he had come to understand he was mistaken when he stated that he "did not participate in the Pharmasset due diligence." Appx19937 (343:13-344:17). "I think I was relying too much on my memory." Appx19937 (344:9-13). Based on the documents, he admitted he must have been on the call. Appx19937 (343:17-25); Appx19938 (347:9-22).

Durette also agreed that, based on the documents, he should have known before the call that PSI-6130 would "be a nucleoside compound." Appx19942 (364:18). But he testified that "nucleosides is a very broad area," and that he and his boss "must have" determined "it was unlikely that there would be an overlap with my HCV docket." Appx19942 (365:16-21). He testified that, according to Roemer's notes, he "told the Pharmasset people that there was an issue" "immediately" after he recognized the problem. Appx19943 (366:19-22).

Durette testified that he took no action on the '499 application between the 2004 call and publication of the Clark application in 2005. Appx19944 (370:4-11); Appx28318-28321. When Clark published, Durette observed, any "obligations under the confidentiality agreement had terminated." Appx19944 (370:13-14). Durette testified that he amended the '499 patent because he felt he would "get an expedition of the examination" if he "narrowed the claim." Appx19945 (376:12-18). But the original claims, he explained, "already encompass[ed] the Pharmasset compound." Appx19944 (370:21-22, 371:25-372:5).

After spending 90 minutes on Durette, *see* Appx19583, Gilead offered only an hour of direct testimony on written description from its lead expert. Appx20146-20152 (729:11-755:24); Appx20202-20205 (776:22-788:10). He devoted just five minutes to the '712 patent. Appx20204-20205 (785:4-788:10).

Gilead's closing invoked Durette 31 times, *see* Appx20660-20666(1658:20-1685:24); included three clips from his deposition, *see* Appx20660(1660:8-1661:5); and devoted at least 10 of its 60 minutes to the March 2004 call and the 2005 amendment to the '499 patent, Appx20660-20662(1658:20-1667:7). Gilead presented no evidence linking the call to the '712 patent. *See* Appx21659 (2608:21-22); Appx00061 n.5.

Merck's case. Merck's case focused on proving that Dr. Olsen and his team discovered the importance of the 2' methyl-up/fluoro-down structure for inhibiting HCV, and that the January 2002 specification adequately described and enabled the asserted claims. Pharmasset, it urged, "looked at our patents and then worked within the scope of our claims" to develop PSI-6130 and later sofosbuvir. Appx19893-19894(169:9-173:23). During opening, Merck told the jury "we don't dispute" that "Durette was on a phone call with Pharmasset in which the structure of 6130 was described." Appx19895(178:5-7). But that call "[wa]s irrelevant" to validity because Merck had possession of its invention earlier. Appx19895(177:7-13).

Merck's lead fact witness, Dr. Olsen, testified for 3½ hours on direct about the development of Merck's inventions. Appx19758. Demain testified about the licensing negotiations with Pharmasset. Appx20498(1391:5). And Merck called three experts on validity. Appx20349(1180:10-11); Appx20476(1306:4-9); Appx20631(1545:14-17).

Merck's closing reiterated that Durette and the March 2004 call were "red herring[s] and really [are] irrelevant" to validity. Appx20676(1723:4-5). If the January 2002 application satisfied enablement and written-description requirements, Gilead's derivation defense—and Durette's actions—would "fall[] out" and become "irrelevant." Appx20677(1726:25-1727:7).

Instructions and Verdict. The district court agreed that the written-description and enablement issues would dispose of Gilead's derivation defense. "If you find that Merck's patent application as filed described and enabled an asserted claim" of the '499 or '712 patents, it instructed the jury, "you must also find that the claim is not invalid for derivation [or] . . . prior invention." Appx19833-19834.

The jury found the claims of both patents not invalid for lack of written description or enablement; it thus did not reach derivation or prior invention. Appx21066-21075. After a damages trial, the jury awarded Merck $200 million for infringement through December 2015. Appx21094-21095.

C. The District Court Overturns the Jury's Verdict, Invoking "Unclean Hands"

The court then held a bench trial on Gilead's equitable defenses, including unclean hands. Appx21607-21608. Relying largely on the testimony before the jury, Gilead urged that Durette's participation in the 2004 due-diligence call, Appx20987, his amendments to the '499 patent, *id.*, and his allegedly "false testimony," Appx21961-21967, were so "unconscionable" as to "bar enforcement of the patents-in-suit," Appx21957; Appx21969.

The court admitted "struggling." Appx21662(2619:4). "I am outraged by what I think was untruthful testimony." Appx22551(17:8). But "I know what egregious conduct the Supreme Court has described as qualifying as unclean hands, and this case does not equal" it. Appx21662(2619:4-8). The court was troubled because Gilead "might prove every fact that you want, and it still doesn't add up to unclean hands." Appx22551(17:11-14). The court emphasized that "it doesn't appear that there's any evidence on the '712" patent. Appx21659 (2608:21-22). The court was "concern[ed]," Appx22550(14:2-4), it could not justify the "enormous punishment" of precluding the '712 patent's enforcement, Appx22551(17:6-7).

The court nonetheless issued an order upsetting the jury's $200 million verdict and "barr[ing]" Merck "from asserting the '499 and '712 Patents against Gilead." Appx00065. The court identified the "trio of cases"—*Keystone Driller Co. v. General Excavator Co.*, 290 U.S. 240 (1933), *Hazel-Atlas Glass Co. v. Hartford-Empire Co.*, 322 U.S. 238 (1944), and *Precision Instrument Manufacturing Co. v. Automotive Maintenance Machinery Co.*, 324 U.S. 806 (1945)—in which "the Supreme Court applied the doctrine of unclean hands to dismiss patent cases involving egregious misconduct." Appx00038-00041. Despite earlier stating that "this case does not equal any of the facts" of those cases, Appx21662(2619:4-8), the court invoked supposed "business" and "litigation" misconduct to foreclose the patents' enforcement.

1. *The '499 Patent*

Business misconduct. The "business misconduct" identified by the court consisted of Durette "learning the confidential structure of Pharmasset compound PSI-6130 and pursuing patent claims to cover that compound in violation of the Merck-Pharmasset firewall and Merck's own policies." Appx00044. Durette, the court stated, should have known before the March 2004 call that Pharmasset's compound would overlap with his prosecution docket, making his participation improper. Appx00022; Appx00024.

The court acknowledged that, during the call, Durette told Pharmasset he had a potential conflict because the subject matter "'seems quite related to things I'm involved with.'" Appx00045 (quoting Appx31545). But the court dismissed that warning because Durette did not specify "he was prosecuting Merck's own HCV patent applications," and he was represented as "within the firewall." *Id.*

The court accused Durette of "improperly us[ing]" information learned on the call "to inform his conduct in amending the '499 Patent." Appx00046. The court did not dispute that Durette did nothing with the ’499 patent until after Clark published PSI-6130, or that Clark’s publication “free[d] [him]” from “his obligations under the NDA.” *Id.* It characterized Durette as “pounc[ing] on the opportunity” when the information became public and thus usable. *Id.*

The court did not acknowledge expressly that Merck’s patent claims ***already*** covered PSI-6130 before the March 2004 due-diligence call and 2005 amendment. It did not mention that Pharmasset knew Merck’s patents covered PSI-6130 before the call. *See* p. 12, *supra*. It acknowledged that the amendments “narrowed” Merck’s claims. Appx00017; Appx00026. But it asserted nonetheless that “Durette would not have written new claims to cover PSI-6130 in February 2005 but for his improper participation” in the March 2004 call. Appx00018.

The court also invoked Durette’s supposed “admi[ssion]” that he “would not have been able to associate any structure in the Pharmasset [Clark] application as . . . PSI-6130 unless he knew the structure of PSI-6130 beforehand.” Appx00018. The court cited deposition testimony in which Durette had been asked about six specific pages of Clark. *See* Appx00018 (citing Appx22345(53:1-6, 53:22-54:5)); *see also* Appx22345-22346(56:2-57:1); Appx23724-23729. The court did not address the fact that, outside those six pages, PSI-6130 was the first Example in Clark and the only new compound with biological data—or that the entire specification and all 130 of Clark's claims recited 2' methyl-up/fluoro-down structures. *See* pp. 15-16, *supra*.

Litigation misconduct. The district court also found "litigation misconduct," which it described as Durette's "inconsistent, contradictory, and untruthful testimony." Appx00047-00053. The court considered it "[r]emarkabl[e]" that, at trial, Durette "recanted a major portion" of his deposition testimony concerning the March 2004 due-diligence call. Appx00047. The court faulted Merck for not "warning" Gilead before opening statements that it would concede Durette was on the call. *Id.* The court did not mention that Gilead had cancelled the depositions of Merck's corporate 30(b)(6) designee (and another witness) about the call, or that Merck produced and identified to Gilead documents showing Durette was on the call. Appx22212-22213(¶8). Nor did it address why Gilead needed "warning" that Merck would agree with Gilead that Durette was on the call.

The court declared that Durette, at his deposition, "did not say that he did not remember a call" or "that he could not be sure" whether he was on the call. Appx00019. The court did not mention Durette's testimony that he "d[id]n't recall such a conversation," Appx22336(19:1-3), or that, when asked whether the call "could . . . have happened and [Durette] just d[id]n't remember," Durette acknowledged: "I guess if I don't remember, both possibilities are possible." Appx22380(194:9-15).

The court did not deny that Durette was a ***former*** Merck employee. But it stated that Merck "fully aligned itself" with Durette by providing him counsel for his deposition and "designat[ing] . . . him as a 30(b)(6) witness on selected issues." Appx00051-00052. The court did not mention that those "selected issues" did not include the 2004 due-diligence call. *See* p. 21, *supra*.

The court faulted Merck for "making Dr. Durette a centerpiece of its case, from the opening statement to the closing argument." Appx00052. It did not mention Merck's effort to exclude his testimony, or that Merck consistently told the jury to ignore Durette's testimony as irrelevant. *See* pp. 23-24, 26-27, *supra*.

No materiality requirement. The court also ruled that unclean hands "does not require a finding of materiality." Appx00041; Appx00060 ("misconduct does not have to be material"). The court recognized that "unclean hands applies only where the 'unconscionable act of one coming for relief has immediate and necessary relation to the equity that he seeks in respect of the matter in litigation.'" Appx00039 (quoting *Keystone*, 290 U.S. at 245). But it interpreted "immediate and necessary" to require only that the misconduct "relate to" the matter before it. Appx00060.

2. *The '712 Patent*

The court recognized that the '712 patent was quite different from the '499. Because the alleged business misconduct had no connection to the '712 patent, the court disavowed reliance on its "finding of improper business conduct" when "determining whether unclean hands prevented enforcement of the '712 Patent." Appx00061 n.5. The court nowhere found that Durette's testimony—the foundation for the alleged litigation misconduct—had any connection to the '712 patent either.

Instead, the court declared that "intentional litigation misconduct casts a darkness on this entire case that covers both patents-in-suit." Appx00060-00061. The '712 patent must be unenforceable, the court stated, because "it would be an odd result . . . if Merck could engage in . . . substantial litigation misconduct . . . yet face no penalty because the '712 Patent was deemed uncontaminated." Appx00061.

D. The District Court Denies Gilead's Post-Trial Motions as Moot

Following the unclean-hands ruling, Gilead moved for judgment as a matter of law, seeking to overturn the jury's validity determinations. Appx21770-21771; Appx33235-33248. Gilead, however, invited the court not to reach those issues because the "determination that the '499 and '712 patents are unenforceable as to Gilead may moot Gilead's request for JMOL." Appx33239 n.1. The court denied Gilead's motion as moot. Appx00066.

SUMMARY OF ARGUMENT

I. A. The district court erred in holding that "unclean hands" bars Merck from enforcing the '499 patent. The exceptional and rare unclean-hands defense bars equitable relief only where a party seeks to reap the benefit of unconscionable misconduct that gave it an unfair advantage or injured its opponent. This Court has called that a "materiality" requirement. Yet the district court held the opposite, declaring that any "materiality requirement" is "non-existent." Under the correct standard, there can be no unclean hands here.

B. The district court purported to find litigation misconduct—namely inconsistent and untruthful testimony by Merck former employee Durette regarding his participation in a March 2004 due-diligence call, and his reasons for amending the '499 patent's claims in 2005. But that testimony was legally irrelevant. Merck had already described, enabled, and claimed the infringing compounds in 2002, so it could not have stolen them from a meeting in 2004; and if Merck had not described and enabled the claims in 2002, the '499 patent was invalid anyway. Moreover, as explained below, by the time of the 2005 amendment, the information in the 2004 call had become public and was open for all to use.

Gilead cannot have been harmed by Durette's statements anyway. At trial, Durette ***agreed*** that he was on the call—and Merck never disputed it. Far from benefitting from the challenged testimony, Merck moved to exclude it; ***Gilead*** insisted that it be put to the jury. ***Gilead*** then focused on the testimony at trial, while Merck told the jury that Durette and the due-diligence call were irrelevant. The notion that Merck sought to reap some benefit from (or Gilead was disadvantaged by) Durette's testimony cannot be reconciled with Merck's efforts to exclude it—or Gilead's efforts to exploit it.

C. The supposed "business misconduct" was immaterial. The court found it wrongful that Durette learned the structure of PSI-6130 in the March 2004 due-diligence call, and supposedly used that information in amending the '499 patent's claims. But Merck's '499 application claimed PSI-6130 years before the due-diligence call. The 2005 amendments only narrowed the claims; they did not give Merck coverage it would otherwise lack. And the amendments occurred after Clark published PSI-6130 for the world to see. As a result, any confidentiality obligations under the NDA had lapsed. This Court's cases make clear that it is permissible to amend claims in light of publicly available information. Merck obtained no unfair advantage from any business misconduct.

D. The district court acknowledged that the alleged misconduct "does not equal" the "egregious conduct the Supreme Court has described as qualifying as unclean hands." Appx21662(2619:4-8). There was no deliberately planned and carefully executed scheme by Merck to defraud. As for litigation misconduct, if

Durette testified falsely—and the court's key findings regarding his testimony are erroneous—there is not a shred of evidence ***Merck*** urged him to so testify. Merck ***agreed*** that Durette was on the March 2004 due-diligence call. And the notion that Merck sought to deceive Gilead, the court, or the jury, is refuted by the fact that Merck moved to ***exclude*** Durette's testimony regarding post-2002 events, and repeatedly told the jury it was irrelevant.

As to the alleged business misconduct, there is no evidence Durette's participation in the due-diligence call was a scheme by Merck to steal Pharmasset's confidential information. Durette expressly told Pharmasset he may have a conflict during the due-diligence call. Moreover, there was no action on the '499 application until after the Clark application published PSI-6130. There is nothing wrongful—under the NDA or otherwise—about using by-then public information to amend claims.

E. Here, "unclean hands" imposed inequity rather than equity. Gilead knowingly infringed Merck's patents. Its predecessor, Pharmasset, had no success developing a novel anti-HCV compound on its own. So it scoured Merck's patent applications to "cash [in] on" Merck's discoveries. Appx24823. Finding no "loopholes," Gilead and Pharmasset infringed anyway. *Id.* Whatever Durette's errors—testifying as a former line-level employee on irrelevant topics where he did not speak for the company—they cannot justify stripping Merck of its patent rights, overturning a $200 million verdict, and licensing Gilead to continue infringing with impunity.

II. The court's ruling on the '712 patent deprives "unclean hands" of any boundaries. The court conceded that the alleged business misconduct did not taint the '712 patent. It identified no way in which Durette's testimony touched on the '712 patent. The court invoked unclean hands for the '712 patent because Merck might not otherwise suffer a "penalty." The desire to punish is not a legitimate basis for precluding recovery on a patent that is valid, infringed, and untainted by any alleged misconduct. Unclean hands cannot extend to the '712 patent.

STANDARD OF REVIEW

The district court's "application of the unclean hands doctrine" is reviewed for abuse of discretion. *Northbay Wellness Grp., Inc. v. Beyries*, 789 F.3d 956, 959 (9th Cir. 2015). A district court necessarily abuses its discretion "'if it does not apply the correct law,'" *id.*, an issue reviewed *de novo*, *Vapor Point LLC v. Moorhead*, 832 F.3d 1343, 1348 (Fed. Cir. 2016). The abuse of discretion standard is also met if the decision "rests . . . on a clearly erroneous finding of material fact." *Northbay*, 789 F.3d at 959 (quotation marks omitted).

ARGUMENT

Unclean hands is an "exceptional" defense. *Polk Bros., Inc. v. Forest City Enters., Inc.*, 776 F.2d 185, 193 (7th Cir. 1985). It is rarely applied to defeat claims for equitable relief, much less "damages and other legal remedies." 1 Dobbs, *Law of Remedies* §2.4(2) (2d ed. 1993). The district court here invoked unclean hands to do just that: It nullified Merck's $200 million verdict and licensed Gilead to continue infringing Merck's patents. The court revoked relief for the '712 patent without finding ***any*** misconduct related to that patent's validity or its assertion. "[O]utraged" by what it considered "untruthful testimony" from a former Merck employee, Appx22551(17:8), the district court denied recovery because, if one patent were found "uncontaminated," Merck would "face no penalty," Appx00061. But unclean hands "does not grant courts free-floating authority to deny" legal rights as punishment. *Cyber Sols. Int'l, LLC v. Pro Mktg. Sales, Inc.*, 634 F. App'x 557, 567-68 (6th Cir. 2016).

Unclean hands ensures that courts do not use their equitable powers to help "a person to reap the benefits of his own misconduct." *Bein v. Heath*, 47 U.S. (6 How.) 228, 247 (1848); *Restatement (Second) of Torts* §940 cmt. c (1979). With respect to the '499 patent as well, that standard (or any other proper unclean-hands standard) was not met. Far from giving Merck an unfair advantage or harming Gilead, the conduct cited by the district court was immaterial. There was no deliberately planned and carefully executed scheme to defraud by Merck. The district court's factfindings often defy the record. And the application of unclean hands here imposes inequity, not equity. The supposed misconduct here cannot remotely support unjustly enriching a knowing infringer by $200 million and granting it a perpetual license to continue infringing.

I. THE '499 PATENT IS NOT UNENFORCEABLE FOR UNCLEAN HANDS

A. The District Court Applied a Legally Incorrect Standard

1. *Unclean Hands Is Limited to Misconduct That Gives the Plaintiff an Unfair Advantage or Injures Its Opponent*

For 150 years, the Supreme Court has emphasized that unclean hands is not a freestanding judicial sanction, but a doctrine that prevents courts from being used to "abet[ ]" an "iniquity." *Bein*, 47 U.S. at 247. In *Kitchen v. Rayburn*, 86 U.S. (19 Wall.) 254 (1874), for example, the Supreme Court invoked the doctrine to bar recovery by parties "seeking the benefit of a contract obtained by their fraud." *Id.* at 263. Unclean hands, the Court stated, precluded the plaintiffs from using the judiciary "to derive an advantage from their own wrong." *Id.* And *Bein* explained that "unclean hands" prevents the "equitable powers of [the] court" from being exploited by one "who by deceit or any unfair means has gained an advantage" with respect to the matters in suit. 47 U.S. at 247.

Although courts have framed that requirement various ways, they have long adhered to its core principle. Because the "rationale" for unclean hands is "that equity will not aid a person to reap the benefits of his own misconduct," it applies only where the "right for which the plaintiff seeks protection . . . accrue[d] to him because of the misdeed." *Restatement*, *supra*, § 940 cmt. c. Some courts focus on injury to the defendant, so that unclean hands does not apply "unless the defendant can show that he has personally been injured" by the wrongdoing. *Dream Games of Ariz., Inc. v. PC Onsite*, 561 F.3d 983, 990 (9th Cir. 2009).[2] Others stress benefit to the plaintiff, declaring that unclean hands prevents plaintiffs from "reap[ing] the benefits of wrongdoing." *In re Uwimana*, 274 F.3d 806, 810-11 (4th Cir. 2001), *abrogated on other grounds*, *Bullock v. Bank-Champaign, N.A.*, 133 S. Ct. 1754 (2013); *see Stiegele v. J.M. Moore Imp.-Exp. Co.*, 312 F.2d 588, 594 (2d Cir. 1963).

This Court has framed the issue in terms of "materiality," calling it "'a necessary ingredient of'" any defense based on "'inequitable conduct,'" including "unclean hands." *Regents of the Univ. of Cal. v. Eli Lilly & Co.*, 119 F.3d 1559, 1571 (Fed. Cir. 1997) (quoting *J.P. Stevens & Co. v. Lex Tex Ltd.*, 747 F.2d 1553, 1560 n.7 (Fed. Cir. 1984)). In *Eli Lilly*, the district court found that the patentee had "falsified material in its patent application." *Id.* at 1569. But this Court reversed the district court's unclean-hands finding because a patent examiner "would not have considered" the alleged misrepresentation "to be material to patentability." *Id.* at 1571.

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[2] *See Bailey v. TitleMax of Ga., Inc.*, 776 F.3d 797, 801 (11th Cir. 2015); *Performance Unlimited, Inc. v. Questar Publishers, Inc.*, 52 F.3d 1373, 1383 (6th Cir. 1995); *Mitchell Bros. Film Grp. v. Cinema Adult Theater*, 604 F.2d 852, 863 (5th Cir. 1979).

However phrased, the law is clear: Unless an advantage or injury accrues "because of" the "misdeed," it is "collateral" and cannot support unclean hands. *Restatement*, *supra*, § 940 cmt. c; *see, e.g.*, *PenneCom B.V. v. Merrill Lynch & Co.*, 372 F.3d 488, 493 (2d Cir. 2004) (New York); *Paramount Aviation Corp. v. Agusta*, 178 F.3d 132, 147 n.12 (3d Cir. 1999) (Pennsylvania); *Alcatel USA, Inc. v. DGI Techs., Inc.*, 166 F.3d 772, 796-97 (5th Cir. 1999) (Texas).

The Supreme Court's application of unclean hands in three 20th-century patent cases—*Keystone Driller Co. v. General Excavator Co.*, 290 U.S. 240 (1933), *Hazel-Atlas Glass Co. v. Hartford-Empire Co.*, 322 U.S. 238 (1944), and *Precision Instrument Manufacturing Co. v. Automotive Maintenance Machinery Co.*, 324 U.S. 806 (1945)—confirms those principles. In *Keystone*, the plaintiff bribed a witness in an earlier suit to suppress evidence of an invalidating prior use, and then invoked its success in that suit when seeking an injunction against the defendant. 290 U.S. at 243, 246-47. Agreeing with the plaintiff's assertion that "the maxim does not apply unless the wrongful conduct is directly connected with and material to the matter in litigation," *id.* at 244, the Court observed that unclean hands does not provide "punishment for extraneous transgressions," *id.* at 245. Instead, it applies "only where some unconscionable act of one coming for relief has immediate and necessary relation to the equity that he seeks in respect of the matter in litigation." *Id.*

The *Keystone* Court concluded that standard was met. Although the misconduct there—suppression of evidence—occurred in a prior lawsuit, the plaintiff had "gained an advantage" in the matter before the Court by using "the decree" wrongfully obtained in that prior action to support "motions for preliminary injunctions" in the current case. 290 U.S. at 246. The Court stressed the "use actually made of that decree" and "the importance of the advantage obtained by [that] use." *Id.* at 247.

Hazel-Atlas and *Precision Instrument* reflect the same understanding. In *Hazel-Atlas*, the patent was "obtained by fraud." 322 U.S. at 239, 251. The plaintiff thus obtained the ultimate advantage—the asserted patent. *Id.* That fraud, the Court held, was "material" to the later infringement suit. *Id.* at 247. And in *Precision Instrument*, the Court held that unclean hands precluded the plaintiff from asserting a patent where the patent application was "based upon false data which destroyed whatever just claim" there might otherwise be "to the status of a patent." 324 U.S. at 816. The Court applied unclean hands to "prevent[] [the] wrongdoer from enjoying the fruits of his transgression" and imposing "an injury to the public." *Id.* at 815. Thus, while courts have articulated the requirements of unclean hands in various ways, the core principle has been consistent—the plaintiff must be seeking to reap the benefit of egregious misconduct that gave it an unfair advantage or injured its opponent in respect of the matter in litigation. Otherwise, the conduct is not material (it is collateral) and not a proper basis for denying the plaintiff relief.

2. *The District Court's Decision Defies Those Settled Principles*

The district court departed from those principles. Any "materiality requirement," the court held, is "non-existent." Appx00060. This Court has held otherwise, requiring "materiality" as "'a necessary ingredient of'" any defense based on "unclean hands." *Eli Lilly*, 119 F.3d at 1571. The district court's decision also defies the purpose of the unclean-hands doctrine—to prevent courts from becoming "abettors of iniquity" by helping the suitor "reap the benefits" of his misconduct.

The district court invoked this Court's decision in *Therasense, Inc. v. Becton, Dickinson & Co.*, 649 F.3d 1276 (Fed. Cir. 2011) (en banc), as "ma[king] clear" that unclean hands "does not require a finding of materiality." Appx00041. It quoted *Therasense*'s statement that the Supreme Court's "'early unclean hands cases do not present ***any standard*** for materiality.'" Appx00041-00042 (quoting 649 F.3d at 1287) (emphasis added). But that reflects only that the Supreme Court has not articulated a "standard" for materiality; it does not mean materiality is not required. Whether or not they call it "materiality," the Supreme Court's unclean-hands patent cases—and countless other cases—plainly require that the wrongdoing confer an advantage on the claimant or injure its opponent in the litigation.

See pp. 39-43, *supra*. Absent that effect, the courts would not be abetting an iniquity in granting the plaintiff relief.

The district court acknowledged that, in *Keystone*, "the Supreme Court explained that unclean hands applies only where the 'unconscionable act of one coming for relief has ***immediate and necessary relation to the equity that he seeks*** in respect of the matter in litigation.'" Appx00039 (quoting *Keystone*, 290 U.S. at 245) (emphasis added). The district court interpreted that to require only that the "misconduct . . . ***relate*** to the asserted claims"—the misconduct, it held, "does not have to be material." Appx00060 (emphasis added). That ignores the Supreme Court's language: "Necessary" means "essential." *Black's Law Dictionary* (3d ed. 1933). Wrongs that neither benefit the wrongdoer seeking relief, nor harm its opponent, cannot be "essential" to "the equity" being sought "in litigation."

Keystone, moreover, did not stop at the "immediate and necessary" formulation. Immediately following that, *Keystone* explains that courts deny relief "because of plaintiff's misconduct" only where it "affect[s] the equitable relations between the parties in respect of something brought before the court for adjudication." 290 U.S. at 245. The district court nowhere explained how misconduct immaterial to the litigation could "affect the equitable relations between the parties" "in respect of" that matter. *Keystone* further explained that unclean hands prevents the court's powers from "be[ing] exerted in behalf of one" who acted fraudulently or by "unfair means has ***gained an advantage***" in the litigation. *Id.* (emphasis added). And it specifically identified the fraudulently obtained advantage in the case before it. *See* pp. 41-42, *supra*. *Keystone* requires more than misconduct that "relates" to the matter before the court.

Ultimately, the district court invoked a statement by Judge Learned Hand that "'the defendant who invokes [unclean hands] need not be damaged.'" Appx00064-00065. That is true, as an unconscionable ***advantage to the plaintiff*** may suffice, even if the defendant suffers no injury. *See* pp. 39-43, *supra*. Regardless, Judge Hand's statement was made in passing, with no citation to authority, in a case where he urged unclean hands should be ***denied***. *See Art Metal Works, Inc. v. Abraham & Straus, Inc.*, 70 F.2d 641, 646 (2d Cir. 1934) (L. Hand, J., dissenting), *dissent adopted on reh'g*, 107 F.2d 944 (2d Cir. 1939). That stray line, even from a renowned judge, cannot override centuries of precedent.

The district court's rejection of any materiality requirement unmoors unclean hands from its purpose. It converts the doctrine from a limited defense in equity into a freestanding basis for punishing litigants, unbound from the requirements for sanctions or contempt. That is particularly inappropriate where unclean hands—an equitable doctrine—is applied to bar a ***legal*** claim for damages. Invoking unclean hands to refuse "damages and other legal remedies" raises the prospect that "citizens would not have rights, only privileges." 1 Dobbs, *supra*,

§ 2.4(2). That "goes too far." *Id.* There is no authority for stripping Merck of its right to enforce a valid patent against an adjudicated infringer based on conduct that affects neither the patent's validity nor the litigation's outcome.

This Court has strived to provide clear standards—including a rigorous materiality requirement—to remedy "the problems created by the expansion and overuse of" the related "inequitable conduct doctrine." *Therasense*, 649 F.3d at 1285. The district court's decision here invites a new "plague" of misconduct allegations, no matter how collateral to the issues in the case, under the rubric of unclean hands. *See id.* at 1289.

B. Merck Gained No Unfair Advantage and Gilead Suffered No Injury from the Alleged "Litigation Misconduct"

Under the correct standard, no finding of unclean hands is possible here. The alleged "litigation misconduct" in no way "affect[ed] the equitable relations between the parties in respect of" the litigation (assuming it occurred at all, *see* pp. 57-61, *infra*). There is no evidence Merck encouraged, controlled, or exploited the challenged testimony—Merck sought to exclude it. Permitting enforcement of the '499 patent would not allow Merck to "gain[ ] an advantage" through "unfair means." *Bein*, 47 U.S. at 247.

The court relied on its finding that "Durette presented inconsistent, contradictory, and untruthful testimony" regarding Gilead's derivation defense—namely, whether Durette participated in the March 2004 call and used information from it to derive Merck's invention. Appx00047. But that testimony—introduced by Gilead not Merck—was legally irrelevant. As Merck had explained, if the January 18, 2002 specification described and enabled the asserted claims, then Merck had conception as of that date. *See Frazer*, 498 F.3d at 1287; Appx15806; Appx21831-21839. Any claim that Merck "derived" its invention from Pharmasset two years later is legally impossible. *See* p. 20 & n.1, *supra*. The jury instructions confirmed that, stating: "If you find that Merck's patent application as filed described and enabled an asserted claim of the '499 patent or the '712 patent, you must also find that the claim is not invalid for derivation [or] . . . prior invention." Appx19833-19834. Conversely, if the original application did ***not*** sufficiently describe and enable the claims, those claims would be invalid anyway and—once again—derivation would be irrelevant. Nowhere did the court explain how Merck could have "gained an advantage" from, *Keystone*, 290 U.S. at 245, or Gilead have "been injured" by, *Dream Games*, 561 F.3d at 990, testimony that was legally irrelevant.

At trial, moreover, Durette ***agreed*** with Gilead that he must have been on the March 2004 due-diligence call and learned of PSI-6130 then. Appx19946-19947(381:24-382:21). The court never explained how testimony favorable to Gilead injured Gilead or helped Merck. And Gilead had ample opportunity to impeach Durette on any perceived deviations from his deposition. *See, e.g.*, Appx19937-19939 (344:8-353:21).

Merck acknowledged—in its opening statement—that "Durette was on a phone call with Pharmasset" in which "6130 was described." Appx19895 (178:5-7). The court faulted Merck for not informing Gilead of that position earlier. Appx00047 & n.3. The court ignored Merck's interrogatory responses, which pointed Gilead to documents showing that "the March 17, 2004 telephone conference regarding PSI-6130" included "Phil Durette." Appx22212-22213 (¶8). It ignored that Merck had designated a different 30(b)(6) corporate witness, ***not Durette***, to testify about the due diligence, including the March 2004 call. Appx22394. But, after deposing Durette, ***Gilead*** "elected to forgo the 30(b)(6) deposition." Appx22396. And the court ignored that ***Gilead*** also cancelled the deposition for Pon—the other Merck participant on the call, Appx22213-22214 (¶¶11-12); *see* pp. 21, 23, *supra*. Merck hid nothing. If anything, Gilead willfully blinded itself to Merck's position.

The only party conceivably injured by Durette's testimony was ***Merck***. The court agreed that Durette's testimony "put[] Merck in a bad light." Appx19413 (79:21). Gilead understood that too—which is why Gilead made Durette a centerpiece of its case:

- ***Gilead*** promised in its opening: "You're going to hear from Mr. Durette ***because we're calling him*** in our case." Appx19885(137:5-6) (emphasis added).
- ***Gilead*** called Durette during its case-in-chief, examining him in detail about the due-diligence call. Appx19937-19943(343:13-369:23).
- ***Gilead's*** closing featured Durette, bringing him up ***31 times*** and featuring ***3 clips*** from his deposition. Appx20660-20666(1658:20-1685:24); Appx20660(1660:8-1661:5).

By contrast, Merck sought to ***exclude*** Durette's testimony as irrelevant and unduly prejudicial. Appx15806-15810; *see* pp. 23-24, *supra*. When that motion was (erroneously) denied, Merck told the jury Durette's testimony is a "red herring and really is irrelevant" to validity. Appx20676(1723:4-5); *see* Appx19895(177:7-13) (Durette testimony "irrelevant"); Appx20677(1726:25-1727:7) (same). The notion that Merck sought to reap some benefit from (or that Gilead was disadvantaged by) Durette's testimony cannot be reconciled with Merck's efforts to exclude it—or Gilead's efforts to exploit it.[3]

C. There Was No Unfair Advantage or Injury from the Alleged "Business Misconduct"

The alleged "unethical business conduct," Appx00044, cannot support unclean hands either. The court found it "unconscionable" that Durette, who was prosecuting Merck HCV patents, "learn[ed] the confidential structure of Pharmasset compound PSI-6130" during the March 2004 due-diligence call, *id.*, and supposedly "used this information to inform his conduct in amending the '499 Patent claims . . . after the Clark application published," Appx00046. The notion that Merck gained some "advantage" from Durette's participation in the call, *Keystone*, 290 U.S. at 245, defies patent law. The '499 patent ***already*** covered PSI-6130 before the call; Durette's amendments ***narrowed*** the claims' scope. And there was no "unfair . . . advantage." *Bein*, 47 U.S. at 247. Durette did nothing to the '499 patent's claims until after PSI-6130 was public. Once that occurred, the information was free for the world to use—including Merck, as the NDA made explicit.

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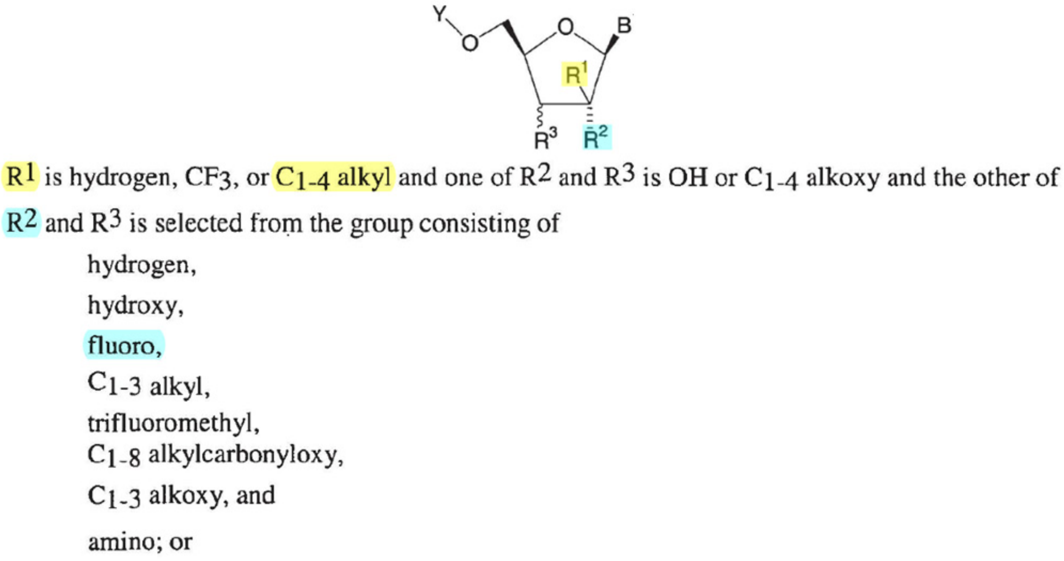
[3] The court stated that Durette "provided key testimony for Merck on validity issues, including written description of the '499 Patent." Appx00023; *see* Appx00052. It identified neither the "key" testimony on that issue nor why it was improper.

1. *The '499 Claims Covered the Relevant Compounds Before the March 2004 Call*

It is undisputed that Merck's '499 application—filed in 2002—claimed PSI-6130 ***before*** the 2004 due-diligence call. In 2003, Pharmasset's Dr. Otto confirmed to colleagues that Merck's patent applications covered PSI-6130, noting "there is methyl up and fluorine down in both the 2' and 3' positions." Appx31458; *see* Appx20051 (545:12-546:12). Dr. Otto thus admitted that, by May 2003, he knew that "[y]ou could find [PSI-6130] in the claims, absolutely." Appx20050 (543:3-20). Gilead's expert, Dr. Secrist, likewise conceded that

Merck's 2002 claims covered "the compound that is 6130." Appx20214(824:7-16).[4]

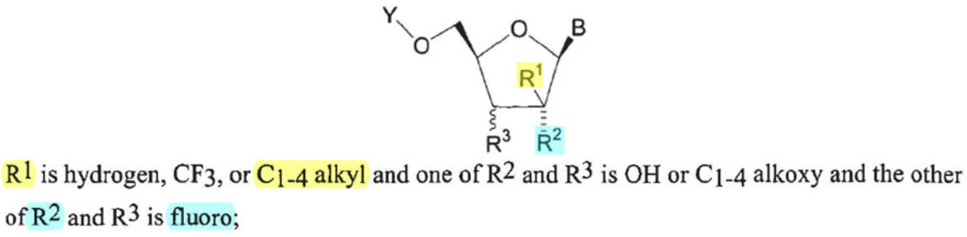
It is also undisputed that the 2005 amendments did not add new compounds to the '499 claims. They instead eliminated candidates for the 2' down (R^2) and 3' (R^3) positions, requiring fluoro at one of those positions. For example, original claim 44 (from July 2003), covers the 2' methyl-up/fluoro-down combination:



R^1 is hydrogen, CF_3, or C_{1-4} alkyl and one of R^2 and R^3 is OH or C_{1-4} alkoxy and the other of R^2 and R^3 is selected from the group consisting of

hydrogen,
hydroxy,
fluoro,
C_{1-3} alkyl,
trifluoromethyl,
C_{1-8} alkylcarbonyloxy,
C_{1-3} alkoxy, and
amino; or

Appx27482-27483 (claim 44, excerpts, colors added). Replacement claim 53 from February 2005 covers the same combination:



R^1 is hydrogen, CF_3, or C_{1-4} alkyl and one of R^2 and R^3 is OH or C_{1-4} alkoxy and the other of R^2 and R^3 is fluoro;

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[4] The court's statement that it "cannot" find "that Pharmasset knew . . . PSI-6130 infringed," Appx00064, is thus baffling.

Appx28319-28321 (claim 53, excerpts, colors added). The difference was elimination of other candidates for the 2' down (R^2) and 3' (R^3) positions (hydrogen, etc.). The court recognized that the amendments did not expand Merck's rights, but "narrowed" the claims. Appx00017; Appx00026.

That forecloses any conclusion of unfair exploitation. The '499 patent's claims covered PSI-6130 before the March 2004 call, and the 2005 amendments narrowed those claims. Merck gained ***no*** new patent rights as a result of Durette's participation in the call. This is not a case where Merck seeks judicial relief to reap the "fruits" of a "transgression." *Precision Instrument*, 324 U.S. at 815. Merck merely seeks the compensation to which the valid '499 patent entitles it.

2. *Merck Also Gained No Unfair Advantage in Amending the '499 Claims Because the Structure of PSI-6130 Was Public*

The court also stated that Durette wrote "new claims to cover PSI-6130 in February 2005," and would not have done so absent "his improper participation" in the March 2004 call. Appx00018. Even apart from the fact that the 2005 amendment only narrowed the claims, there was nothing "unfair" or "wrongful" about it. It occurred only ***after*** the Clark application published PSI-6130's structure for all to see in 2005. The law and the parties' agreement made clear that Merck was free to use such public information.

It is not "in any manner improper" for a patent applicant to broaden its claims to cover a competitor's publicly disclosed product where—as the jury found here—the original disclosure encompasses the amendment. *Kingsdown Med. Consultants v. Hollister, Inc.*, 863 F.2d 867, 874 (Fed. Cir. 1988). *A fortiori*, Durette gained no unfair advantage by narrowing the '499 patent's claims, which already covered the relevant compounds beforehand.

The district court responded that "*Kingsdown* does not permit individuals to disregard firewalls and confidentiality agreements." Appx00057. But the court found no violation of the NDA—and with reason. The NDA provided that information that "becomes part of the public domain" ceases to be "[c]onfidential [i]nformation" subject to its restrictions. Appx32152(¶3(ii)). Here, Durette took no substantive action on the '499 application until after Clark published. *See* pp. 16-18, *supra*. That publication undisputedly put the information in the public domain. As Gilead's counsel urged, once Clark "published in January 2005," "***now the whole world can see the structure*** .... ***There's 6130***." Appx19886(144:7-12) (emphasis added). Durette thus understood that, once Clark published, PSI-6130 was no longer confidential, and he was "free from ... his obligations under the NDA." Appx00046. While the court described Durette as "lying in wait," Appx21644(2546:10-11), that is just a pejorative way of saying he took no action until confidentiality restrictions expired.

The court also suggested that Durette violated a "firewall." Appx00044. But it cited no evidence that the firewall's duration exceeded the NDA's. To the contrary, Roemer described the due-diligence call as "privileged . . . under our [N]DA under the ***firewall of the agreement***." Appx31545 (emphasis added). And the court found that the firewall's "purpose . . . was to protect Pharmasset's confidential structural information." Appx00011-00012. The firewall had no purpose once the structural information became public.

Nor did "Merck's own policies," Appx00044, suggest any unfair benefit. Under (an apparently unwritten) Merck policy, licensing-related due diligence would not be assigned to an attorney prosecuting a "docket in a related area," because "potential issues" might create an "appearance of impropriety." Appx22341 (39:3-40:4). Durette thus admitted that, in hindsight, "[i]t was a mistake" for him to be on the due-diligence call. Appx19943 (366:9). But Durette did not use confidential information from the call. He acted only after the information was public. It was ***not*** against Merck policy to use published information in prosecution. Appx22380 (195:21-24). Thus, while violation of an internal policy is not the stuff of unclean hands, Durette's '499 patent amendments violated no policy in any event.

According to the court, Durette "admitted that he would not have been able to associate any structure in the [Clark] application as the structure of PSI-6130 unless he knew the structure of PSI-6130 beforehand." Appx00018; *see* Appx00021; Appx00026. That defies Gilead's concession that, once Clark published, "the whole world c[ould] see . . . 6130." Appx19886(144:7-12). It defies the record. Shown ***only six pages*** of the Clark application in his deposition, Durette said he would not have associated the structures ***in those pages*** with PSI-6130. *See* Appx22345(53:1-6, 53:22-54:5); Appx22345-22346(56:2-57:1); Appx23724-23729. And it defies Clark's content, which highlights PSI-6130. Clark's Examples described routes for synthesizing only three compounds; both Examples 1 and 2 describe PSI-6130. Appx27074. PSI-6130 was the ***only*** new compound for which biological data was reported. Appx27085-27092. And, as Durette testified, that data showed PSI-6130 was "very active." Appx19948-19949(389:23-390:14); *see also* pp. 15-16, *supra*.

Even if PSI-6130 were somehow hidden, Clark was ***all*** about 2' methyl-up/fluoro-down nucleosides. It described "the present invention" as 2' methyl-up/fluoro-down nucleosides. Appx26999; Appx27014; Appx27023. All 130 claims recited 2' methyl-up/fluoro-down structures. *See* Appx27093-27221. All nucleosides depicted in the specification (except intermediate steps) have a 2' methyl/fluoro structure. Appx27029-27043. And the Example compounds are all 2' methyl-up/fluoro-down nucleosides. Appx27074-27085. The notion that anyone could have missed the publication's focus on 2' methyl-up/fluoro-down nucleosides defies credulity.

D. The Findings Do Not Establish the Egregious Misconduct Required for Unclean Hands

The record, moreover, does not evidence the "egregious misconduct" necessary for unclean hands. *Therasense*, 649 F.3d at 1287. The Supreme Court's "trilogy" all involved a plaintiff's "'deliberately planned and carefully executed scheme to defraud'" the PTO and the courts. *Id.* In *Keystone*, the patentee paid a witness to falsify an affidavit and suppress evidence. 290 U.S. at 243-44. In *Precision Instrument*, the inventor secured a patent through "perjury," and the patent owner "suppressed facts" to conceal that from the PTO and the courts. 324 U.S. at 818-19. And, in *Hazel-Atlas*, the patentee induced the court to rely on an article he had paid an expert to produce for the "deliberate purpose of deceiving the Patent Office." 322 U.S. at 250.

The district court acknowledged the level of "egregious conduct the Supreme Court has described as qualifying as unclean hands," and confessed that "this case does not equal any of the facts in *Keystone* or *Hazel-Atlas* or *Precision*." Appx21662(2619:4-8). That was right. The court was incensed by what it thought was false deposition testimony from Durette—a former line-level employee on a topic where he did not speak for Merck. Appx00047. But ***Merck*** consistently agreed that Durette participated in the March 2004 due-diligence call and learned of PSI-6130. Merck never hid that. And while the court relied on Durette's amendment to the '499 patent's claims, neither the amendment nor testimony about it was wrongful.

1. *There Was No Egregious Litigation Misconduct*

No plan to defraud or deceive. Whatever one might say of Durette's deposition testimony, it does not suggest Merck had a "'deliberately planned and carefully executed scheme to defraud.'" *Therasense*, 649 F.3d at 1287. Indeed, there is no evidence Merck intended to mislead at all, much less that deception was "the 'single most reasonable inference to be drawn from the evidence.'" Appx00057. Durette was a former employee. He was not Merck's 30(b)(6) witness about the due-diligence call. Merck thus could not control Durette, and Durette could not "speak[] for the corporation" on that topic. 8A C. Wright & A. Miller, *Fed. Prac. & Proc.* §2103 (3d ed.). While the court stated that Merck "aligned itself" with Durette by providing him "legal counsel," and designating him a 30(b)(6) witness on other, non-due-diligence topics, Appx00051, it conspicuously did ***not*** find that ***Merck*** urged Durette to testify falsely. That alone distinguishes this from the Supreme Court's trilogy.

Merck never hid Durette's participation on the call. It produced documents showing his participation, highlighted them in its interrogatory answers, and admitted his participation in opening. *See* pp. 21, 26, *supra*. After Durette's deposition, ***Gilead*** cancelled the depositions of ***Merck's designated witness*** on the due-diligence call and of the only other Merck participant. *See* pp. 23, 48, *supra*. ***Gilead*** seems to have executed a "deliberately planned scheme" to avoid information beyond Durette's deposition.

Merck did the opposite of exploiting Durette's supposedly false testimony. Merck moved to exclude that evidence. *See* Appx15720-15726; Appx15804-15810; pp. 23-24, *supra*. ***Gilead*** demanded that it be put to the jury. Likewise, Merck told the jury that Durette's testimony about the due-diligence call was "irrelevant to this case," Appx19895(177:7-13), that these issues were "red herring[s] and really [were] irrelevant," Appx20676(1723:4-5). And Merck ***agreed*** with Gilead that "[Durette] did attend the meeting and he did learn the structure of 6130. There's no dispute." Appx20676(1723:18-19) (closing); *see* Appx19895(178:5-7) (opening).

Far from making supposedly tainted testimony "a centerpiece of its case, from the opening statement to the closing argument," Appx00052, Merck sought to exclude it. Gilead pressed it. If Durette gave false testimony, there is no evidence it was part of a "'deliberately planned and carefully executed scheme [by Merck] to defraud'" Gilead or the court. *Therasense*, 649 F.3d at 1287.

Clear error regarding Durette's testimony. The court's characterization of Durette's testimony also constitutes clear error. The court castigated Durette for having "definitively stated [during his deposition] that he was sure he was never on the call." Appx00019. Durette, it declared, "***did not say*** that he ***did not remember*** a call or that he ***could not be sure***." *Id.* (emphasis added). But Durette said precisely that. He repeatedly said he "d[id]n't ***recall*** such a conversation." Appx22336(19:1-3) (emphasis added); *see* Appx22374-22375(172:22-174:21); pp. 21-22, *supra*. Indeed, at one point Gilead's counsel declared: "***I know you don't remember it.*** What I'm asking you is: . . . [C]ould [the call] have happened and ***you just don't remember***?" Appx22380(194:9-11) (emphasis added). Far from "definitively" denying participation, Durette responded, "I guess if I don't remember, both possibilities are possible." Appx22380(194:14-15). The court's inaccurate finding about Durette's testimony—adopted nearly verbatim from Gilead's proposed factual findings, *see* Appx22090—defies the record.

To be sure, elsewhere in his deposition, Durette declared that he "never participated in a due diligence meeting." Appx22341(37:13-18); *see* Appx22339 (30:21-31:10) ("positive" he "never saw" PSI-6130 until Clark published). But that was right after he admitted his memory was "not very strong." Appx22340(34:24-35:5). And his testimony ended where it began—saying he did not remember. Appx22380(194:1-15).

Finally, the court faulted Durette for trial testimony that he had come to know he had "participate[d] at the meeting." Appx19938(347:16-22). The court called it "[r]emarkabl[e]" that "Durette recanted his testimony that he had not been on the Pharmasset-Merck due diligence call." Appx00047. But Durette's trial testimony was not that he suddenly remembered. It was that—according to documents he reviewed—he must have been on the call. Appx19937(343:17-25); Appx19938(348:17-349:1). Besides, the court never explained how Durette's agreement with Gilead that he participated in the call—testimony that was correct—could constitute egregious litigation misconduct.

The Clark application's role in the 2005 amendments. At his deposition, Durette testified that he did not recall when he first saw the Clark application. Appx00049-00050; *see* Appx22344(51:25-52:1). The court accused Durette of an about-face—that, at trial, he "recalled clearly that he did see the Clark publication before he filed the amendments." Appx00050. That was not Durette's trial testimony. Durette still testified that he ***lacked*** "a specific recollection of the timing." Appx19949(390:25). Durette merely surmised that he "must have" seen Clark before filing the amendment, as it was the likely "triggering event" for him revisiting the dormant '499 docket. Appx19949(390:25-391:9). Moreover, any perceived "inconsistencies," Appx00051, are beside the point. Durette agreed with Gilead's position at trial—that he must have seen Clark before submitting the amendments. So did Merck. *See* Appx20676(1725:4-18). That is not wrongful, much less egregious misconduct.

Finally, the court faulted Durette for having "offered different reasons" for amending the '499 patent's claims. Appx00050. At both trial and his deposition, Durette said he hoped narrowing the claims to focus on 2' or 3' fluoro would "expedite prosecution of the application." Appx22347(62:8-9) (deposition); Appx19945(376:12-18) (trial). At trial, Durette also stated that he "narrowed the claims" to require fluoro at the 2' or 3' position "because we wanted to get an allowance on the subject matter that was most important to the [Merck-Isis] collaboration"—matter "for which [Merck] already had support in the specification" and that "was already . . . claimed." Appx19952(404:15-19). The court found that "not credible because Merck never tested any of the claimed compounds." Appx00027. But that ignores years of research at Merck, including Dr. Olsen's "important finding" that compounds featuring 2' fluoro had properties critical to chain terminators for the HCV polymerase, Appx20299(980:10-982:2), and that he "made sure" they were included in the patent's formula and general claims, Appx20297(973:1-975:16); *see also* Appx25818-25819; Appx25954-25956; pp. 8-9, *supra*.

2. *The Business Misconduct Finding Cannot Be Sustained*

The district court's finding of "unconscionable" business misconduct likewise falls short. The court declared it was "improper" for Durette to participate in the March 2004 due-diligence call because its subject overlapped with his prosecution docket. Appx00014. But there is reason to believe Durette's participation in the one call was at most a mistake, not a deliberate plan. Merck's '499 application, which covered PSI-6130, was public by July 2002. Appx24832. Durette had no reason to expect Pharmasset would be trying to license Merck a compound that, in Dr. Schinazi's words, Merck "already had in their stable" under the '499 patent. Appx20341-20342(1151:11-1152:1). As Merck's Doug Pon told Pharmasset, "when we started the negotiations, we assume[d] that [PSI-6130] was novel." Appx32188. And when Durette, at the outset of the call, specifically asked whether the compound was covered by non-Pharmasset patents—including Merck's—Pharmasset's representatives said nothing. *See* pp. 13-14, *supra*.[5]

By contrast, when Durette realized there might be overlap with his docket, he did not remain silent. He announced on the call that "[i]t's a problem," it "'seems quite related to things that I'm involved with.'" Appx31545. The court stated that Durette waited until "after learning key structural features of PSI-6130" to raise his potential conflict. Appx00045. But Roemer's notes show that Durette raised it before the "[c]ompound [was] described." Appx31545. Roemer's notes reflect that, at another time during the meeting, it was represented that Durette was "within the firewall." *Id.* However, Durette explicitly told Pharmasset he was "conflicted." *Id.* Durette's disclosure of a potential conflict refutes the notion that he, or Merck, was pursuing a calculated plan to deceive.

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[5] Faulting Merck's counsel, the court also declared: "Merck's argument that it openly acknowledged Dr. Durette's participation in the 2004 phone call overlooks that in the very next sentence, its counsel told the jury that Dr. Durette appeared on the phone call because he did not know the compound that was going to be disclosed was within the scope of the Merck patent applications he was working on which turned out to be false." Appx00059; *see also* Appx00022. It is true that PSI-6130 was within Merck's patent applications. But there is no evidence that Dr. Durette or anyone at Merck knew that before the call.

The court's claim that Merck committed misconduct in "pursuing patent claims to cover [PSI-6130] in violation of the Merck-Pharmasset firewall and Merck's own policies," fails as well. Appx00044. PSI-6130 was already covered before the March 2004 call. And Durette took no action on the '499 patent until after Clark published PSI-6130, removing any prohibition on use. Lawful use of public-domain information as permitted by the terms of the NDA and this Court's precedent hardly amounts to a "deliberately planned and carefully executed scheme to defraud."

E. The Equities Favor Enforcing the '499 Patent

"[D]etermining whether the doctrine of unclean hands precludes relief requires balancing the alleged wrongdoing of the plaintiff against that of the defendant, and 'weigh[ing] the substance of the right asserted by [the] plaintiff against the transgression which, it is contended, serves to foreclose that right.' " *Northbay*, 789 F.3d at 960 (quoting *Republic Molding Corp. v. B.W. Photo Utils.*, 319 F.2d 347, 350 (9th Cir. 1963)) (alterations in original); *see Johnson v. Yellow Cab*

Transit Co., 321 U.S. 383, 387 (1944). The district court failed to do that. Its analysis was limited to the passing statement that patent infringement "is serious." Appx00063. The requirement that the court "'weigh[] the substance'" of infringement against Durette's supposed "'transgression[s]'" demands more.

Merck spent years testing over 2,000 nucleoside analogs to identify a class of compounds—including 2' methyl-up/fluoro-down structures—active against HCV. *See* pp. 4-5, *supra*. Before seeing Merck's work, Pharmasset spent years without making a single new compound active against HCV. *See* pp. 10-12, *supra*. It was Merck's work that provided Pharmasset's "rationale" for pursuing compounds like PSI-6130. Appx31499; Appx31507 n.35; Appx31084. Jeremy Clark had a copy of Merck's patent application ***in his hand*** when he told Dr. Otto his ideas for potentially active compounds. Appx20040 (502:8-503:9); Appx20048 (533:11-534:1).

Gilead then intentionally infringed. Dr. Otto admitted that, in 2003, Pharmasset "knew" PSI-6130 "was claimed by the [Merck] patent." Appx20050 (543:13-14). But Pharmasset developed PSI-6130, and Gilead later marketed Sovaldi® and Harvoni®, without a license anyway. Gilead and Pharmasset should reap benefits from their contributions to HCV treatment. But they cannot "look[] at [Merck's] patents and then work[] within the scope of [Merck's] claims" without acquiring a license. Appx19893-19894 (169:9-175:5).

It would be inequitable for Merck to be denied recompense for Gilead's intentional infringement. It is doubly inequitable to deny Merck its rights based on a call in 2004, years after Merck's invention. Having heard all the evidence Gilead presented on the due-diligence call (including its cross-examination of Durette), the jury concluded that Merck had described and enabled claims covering 2' methyl-up/fluoro-down compounds in 2002, long before the due-diligence call, and thus could not have derived them from Gilead. *See* p. 27, *supra*. The court was required to accept those findings. *See Cabinet Vision v. Cabnetware*, 129 F.3d 595, 600 (Fed. Cir. 1997) (Seventh Amendment constrains judicial factfinding where there is "'substantial commonality'" between the facts underlying an inequitable-conduct issue and jury issues). Durette's narrowing of the claims in 2005, after PSI-6130 was published and any confidentiality restrictions expired, does not change the equities. The jury's findings and the testimony of Pharmasset's own witnesses show that Pharmasset (and Gilead) unlawfully took the fruits of Merck's labor, not the other way around.

II. THE '712 PATENT IS NOT UNENFORCEABLE FOR UNCLEAN HANDS

Wholly apart from the '499 patent, the district court's ruling on the '712 defies all boundaries on the unclean-hands doctrine. The court conceded that the alleged business misconduct did not taint the '712 patent. And it identified no way in which Durette's testimony touched on the '712 patent. Instead, the court invoked unclean hands for the '712 patent because Merck might not otherwise suffer a "penalty" for other conduct. The desire to ensure a "penalty" is not a legitimate basis for precluding enforcement of an untainted patent.

A. The District Court's Rationale for Applying Unclean Hands to the Untainted '712 Patent Exceeds the Doctrine's Scope

Throughout the hearing on unclean hands, the court observed that "it doesn't appear that there's any evidence" regarding "the '712." Appx21659(2608:21-22). Gilead's attempt to "trace" prosecution of the '712's claims "back to Durette was a dead end." Appx21648(2561:20-21). And Durette's alleged conduct, the court observed, did not "taint the litigation to the level necessary." Appx22551(17:18-19). Simply put, nothing in Gilead's unclean-hands case had any connection to the '712 patent.

The '712 patent was in a separate family: Filed in 2007, it did not claim priority to the PCT application that issued as the '499 patent; its parent was a separate, non-provisional application also filed on January 18, 2002. *See* Appx00223. Durette had no involvement in the '712 patent's prosecution beyond the initial filing. He retired in June 2010, at a time when the '712 patent's claims did not cover the accused products. Appx19954(413:18-23). In 2010, Pharmasset publicly disclosed sofosbuvir. Appx31990-32007. In 2011, a different patent prosecutor (Jeffrey Bergman) added claims to the '712 patent to cover sofosbuvir's metabolites. *See* pp. 18-19, *supra*. But he had no connection to the 2004 due diligence.

The court thus disavowed relying on its "finding of improper business conduct . . . in determining whether unclean hands prevented enforcement of the '712 Patent." Appx00061 n.5. It identified no allegedly false or inconsistent Durette testimony bearing on the '712 patent. Instead, "outraged" by Durette's testimony on ***other*** issues, Appx22551(17:8), and concerned that Gilead might "win the battle and lose the war" if the '712 patent were "deemed uncontaminated," Appx22551(17:11-12); Appx00061, the court invoked a different rationale: Applying unclean hands was necessary because Merck otherwise would suffer "no penalty" for "substantial litigation misconduct" in connection with the '499 patent. Appx00061; *see* Appx22551(17:6-7) ("enormous punishment for unclean hands").

That is error. Unclean hands is "not a source of power to punish." *Aptix Corp. v. Quickturn Design Sys., Inc.*, 269 F.3d 1369, 1378 (Fed. Cir. 2001); *see also Johnson*, 321 U.S. at 387. True perjury, of course, would be reason for judicial umbrage and potential sanctions for those suborning it. But invoking unclean hands as a "penalty" unmoors it from historical justification. It converts a doctrine designed to prevent courts from becoming an "abetter of iniquity," *Bein*, 47 U.S. at 247, into an auxiliary sanctions provision—one that precludes enforcement of "uncontaminated" patents based on subjective "outrage."

B. The District Court's Pall of "Darkness" Rationale Fails

Unable to link the '712 patent to the supposed business misconduct, the court resorted to metaphor: The litigation misconduct, it asserted, "casts a darkness on this entire case that covers both patents-in-suit." Appx00060-00061. That applies no recognized unclean-hands standard. It ignores materiality, identifying no way in which Merck obtained an advantage, or Gilead was injured, with respect to the '499 patent, much less the '712. And Durette's testimony—or any "darkness" emanating from it—was injected into the trial by Gilead, not Merck. The contention that Merck made Durette "a centerpiece of its case, from the opening statement to the closing argument," Appx00052, is backward: Gilead made him a centerpiece, and Merck dismissed him as "irrelevant." *See* pp. 24-27, *supra*. Likewise, the court's assertion that Durette's testimony "supported Merck's validity arguments, and went to the heart of significant issues," Appx00052, is unsupported. The testimony the court challenged had no bearing on either patent's validity, as the court's own jury instructions made clear. *See* p. 27, *supra*. And the court never explained what "significant issue" regarding the '712 Durette's testimony "went to."

Indeed, the court did not explain how Durette's testimony touched on ***any*** issue concerning the '712 patent. Even under the court's (erroneous) view of the "immediate and necessary relation" standard, misconduct still must "***relate*** to the asserted claims." Appx00060 (emphasis added). No relationship to the '712 patent was shown at all.

The court's analysis thus reduced to the happenstance that Gilead brought a declaratory judgment action concerning the '712 and '499 patents in a single lawsuit. *See* Appx00061. But the court had no authority to wipe out a $200 million jury verdict, independently supported by the untainted '712 patent, because of alleged misconduct with no relationship to that patent. Even where one patent proves tainted by misconduct, that does not defeat claims under another patent simply because they were "brought . . . in the same lawsuit." *Hoffman-La Roche, Inc. v. Promega Corp.*, 319 F. Supp. 2d 1011, 1026 (N.D. Cal. 2004). Unclean hands provides that "wrongdoing directly connected ***with the right sought to be vindicated***" may "deprive one of a remedy" with respect to "***that property right***." *Maatschappij Tot Exploitatie Van Rademaker's Koninklijke Cacao & Chocoladefadrieken v. Kosloff*, 45 F.2d 94, 96 (2d Cir. 1930) (emphasis added). But "it does not outlaw the wrongdoer and leave him without protection in the vindication of other, though closely connected, rights." *Id.* It does not "abet" an "iniquity" for a court to enforce a valid patent against an adjudicated infringer, whatever the purported misconduct with respect to other patents. *Bein*, 47 U.S. at 247.

Keystone is instructive. The respondents there argued that a court may dismiss an "entire" lawsuit, "whether unclean hands [is] applied to one patent or all patents." Resp. Br. in *Keystone Driller Co. v. Gen. Excavator Co.*, Nos. 34, 35, 36, 37, 1933 WL 31881, at *9 (U.S. filed Oct. 16, 1933). Declining to embrace that expansive theory, the Court barred enforcement of five patents only after determining that "the plaintiff's unclean hands had a direct effect on ***each*** of the causes of action . . . for ***each*** of the separate patents asserted." *Consol. Aluminum Corp. v. Foseco Int'l Ltd.*, 910 F.2d 804, 810 (Fed. Cir. 1990) (emphasis added); *see Keystone*, 290 U.S. at 247 ("plaintiff did not come with clean hands in respect of ***any*** cause of action" (emphasis added)); pp. 41-42, *supra*.

The court failed to engage in that patent-by-patent analysis here. It had no power to bar the '712 patent's enforcement on the ground that it was acceptable collateral damage to ensure a "penalty." Given the absence of any misconduct connected to the '712 patent, that approach leaves "unclean hands" with virtually no boundaries.

CONCLUSION

The district court's judgment should be reversed.

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Respectfully submitted,

/s/ Jeffrey A. Lamken

Jeffrey A. Lamken
Counsel of Record
Michael G. Pattillo, Jr.
Sarah J. Newman
James A. Barta
MOLOLAMKEN LLP
The Watergate, Suite 660
600 New Hampshire Ave., N.W.
Washington, D.C. 20037
(202) 556-2000 (telephone)
(202) 556-2001 (facsimile)
jlamken@mololamken.com

Sara Margolis
MOLOLAMKEN LLP
430 Park Avenue, Floor 6
New York, NY 10022
(212) 607-8160 (telephone)
(212) 607-8161 (facsimile)

Bruce R. Genderson
Jessamyn S. Berniker
Stanley E. Fisher
Sanjiv P. Laud
Jessica Palmer Ryen
WILLIAMS & CONNOLLY LLP
725 Twelfth Street, N.W.
Washington, D.C. 20005
(202) 434-5000 (telephone)
(202) 434-5029 (facsimile)
bgenderson@wc.com

Stephen S. Rabinowitz
Mitchell Epner
HUGHES HUBBARD & REED LLP
One Battery Park Plaza
New York, NY 10004
(212) 837-6000 (telephone)
(212) 299-6807 (facsimile)
stephen.rabinowitz@hugheshubbard.com

Counsel for Merck & Co., Inc., Merck Sharp & Dohme Corp., and Isis Pharmaceuticals, Inc.

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UNITED STATES DISTRICT COURT

NORTHERN DISTRICT OF CALIFORNIA

SAN JOSE DIVISION

GILEAD SCIENCES, INC., Plaintiff, v. MERCK & CO, INC., et al., Defendants.	Case No. 13-cv-04057-BLF **ORDER REGARDING NON-JURY LEGAL ISSUES** [Re: ECF 407, 411]

Plaintiff Gilead Sciences, Inc. ("Gilead") seeks to bar Defendants Merck & Co., Merck Sharp and Dohme Corp., and Isis Pharmaceuticals, Inc., (collectively "Merck") from maintaining their suit based on the equitable defenses of waiver and unclean hands. At trial, the jury determined that Merck's patents-in-suit are not invalid and awarded damages to Merck for infringement. Gilead's equitable defenses, however, are the province of the Court to decide.

After a thorough review of the evidence submitted at trial and in post-trial submissions, the Court finds Gilead has not shown that Merck waived its right to enforce the '499 and '712 Patents against Gilead. The record, however, reflects a pervasive pattern of misconduct by Merck and its agents constituting unclean hands, which renders Merck's '499 and '712 Patents unenforceable against Gilead.

I. BACKGROUND

On December 6, 2013, Gilead received approval from the Food and Drug Administration to market and sell Sovaldi®, an orally-administered prescription drug containing the active ingredient sofosbuvir, to treat chronic Hepatitis C (HCV) infection in patients. Order Construing Claims at 2, ECF 140. Sofosbuvir is a prodrug that is inactive and has little to no therapeutic effect until transformed by enzymes in the body into an active form. *Id*. Once inside a liver cell, sofosbuvir is converted into three analogs, each with different structures: a monophosphate analog, a diphosphate analog, and a triphosphate analog. *Id*. The triphosphate analog is the therapeutically effective form that can target and cure HCV infection in patients. *Id*.

Merck asserts that two of its patents, U.S. Patent No. 7,105,499 and U.S. Patent No. 8,481,712, cover sofosbuvir, and that Gilead's sales of Sovaldi® and Harvoni®, which contain the active ingredient sofosbuvir, induce and contribute to the infringement of these patents. Merck Mot. for SJ, ECF 167. The operative filing date of the '499 and '712 Patents is January 18, 2002. Exh. 22 to Gilead Mot. for SJ at Interrog. No. 1, ECF 164-16.

The '712 Patent is directed to compounds having a specific structural formula, Exh. 16 to Gilead Mot. for SJ at 143:1-146:60, ECF 165-11, while the '499 Patent relates to methods for treating HCV by administering a therapeutically effective amount of those compounds either alone or in combination with another HCV treatment. Exh. 1 to Gilead Mot. for SJ at 137:1-138:25 (claims 1 and 2).

At summary judgment, Gilead argued that the asserted claims were invalid but conceded that if they were not invalid, it infringed them. The Court denied Gilead's summary judgment motion of invalidity and granted Merck summary judgment of infringement. ECF 214. On March 20, 2016, after an eight-day trial, the jury found that the '499 and '712 Patents were not invalid. Following a three-day trial on damages, the jury awarded Merck $200 million in damages for sales of Sovaldi® and Harvoni® through December 31, 2015. Verdict Phase 2, ECF 392. On March 30, 2016, the Court held a bench trial on Gilead's equitable defenses of unclean hands and waiver. ECF 401. On April 22, 2016, Gilead filed a motion to re-open the record and allow additional evidence. ECF 410. On April 29, 2016, the Court held a hearing on Gilead's motion where the Court granted the motion and also allowed Merck to supplement the record. ECF 418.

II. LEGAL STANDARD

Federal Rule of Civil Procedure 52(a) requires district courts to make findings of fact in an action "tried on the facts without a jury or with an advisory jury." Fed. R. Civ. P. 52(a)(1). The Court is required to "find facts specially and state its conclusions of law separately." *Id*. "One purpose behind Rule 52(a) is to aid the appellate court's understanding of the bases of the trial

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court's decision." *Simeonoff v. Hener*, 249 F.3d 883, 891 (9th Cir. 2001) (internal citations omitted). The Court is not required to make findings on each and every fact presented at trial. *Id*. Conflicting testimony must be resolved on relevant issues. *Zivkovic v. Southern California Edison, Co.*, 302 F.3d 1080, 1090 (9th Cir. 2002).

III. FINDINGS OF FACT

Gilead argues that Merck waived its rights to enforce the '499 and '712 Patents, or alternatively, that these patents are unenforceable by virtue of the doctrine of unclean hands. Gilead Trial Br., ECF 368; Gilead Supp. Trial Br., ECF 408. Gilead claims Merck impliedly waived its patent rights by attempting to license or acquire from Pharmasset, Gilead's predecessor-in-interest, its confidential compound, PSI-6130 from 2003 to 2011. Gilead Trial Br. 8-9, ECF 368. Next, Gilead argues Merck's unclean hands bars enforcement of the patents against it because Merck improperly obtained the structure of PSI-6130 from Pharmasset, drafted patent claims covering PSI-6130, and then lied about its conduct during this proceeding. Gilead Trial Br. 2-8, ECF 368. Merck responds that it never explicitly or implicitly indicated that it would not enforce the '499 and '712 Patents against Gilead. Merck Tr. Br. 5-6, ECF 370. Merck also argues the jury's rejection of Gilead's invalidity defense forecloses Gilead's unclean hands defense and even if it did not, Merck's actions do not warrant a finding of unclean hands. Merck Trial Br. 1-6, ECF 370; Merck Supp. Trial Br., ECF 409. With that brief overview of the parties' arguments, the Court makes the following findings of fact and conclusions of law.[1]

A. The Parties

1. Plaintiff Gilead Sciences, Inc. ("Plaintiff" or "Gilead") and Defendants Merck & Co., Inc. ("Merck & Co."), Merck Sharp & Dohme Corp. ("MSD Corp."), and Ionis Pharmaceuticals, Inc., formerly known as Isis Pharmaceuticals, Inc. ("Ionis" or "Isis"), (collectively, "Defendants" or "Merck") are the parties in this action. Compl., ECF 1.

2. Gilead is a company organized and existing under the laws of the State of Delaware with its principal place of business at 333 Lakeside Drive, Foster City, California 94404. Compl.

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[1] To the extent that any conclusion of law is deemed to be a finding of fact, it is adopted as such; and likewise, any finding of fact that is deemed to be a conclusion of law is so adopted.

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¶ 2, ECF 1.

3. Merck & Co. is a company organized under the laws of the State of New Jersey with its principal place of business at One Merck Drive, P.O. Box 100, Whitehouse Station, NJ 08889-0100. Compl. ¶ 3, ECF 1; Ans. ¶ 3, ECF 62.

4. MSD Corp. is a company organized under the laws of the State of New Jersey with its principal place of business at One Merck Drive, P.O. Box 100, Whitehouse Station, NJ 08889-0100. Compl. ¶ 4, ECF 1; Ans. ¶ 4, ECF 62.

5. MSD Corp. is a subsidiary of Merck & Co. Compl. ¶ 5, ECF 1; Ans. ¶ 5, ECF 62.

6. Ionis is a company organized under the laws of the State of Delaware with its principal place of business at 2855 Gazelle Court, Carlsbad, CA 92010. Compl. ¶ 6, ECF 1; Ans. ¶ 6, ECF 62.

B. General Background of the Litigation

7. The patents-in-suit are U.S. Patent Nos. 7,105,499 (the "'499 Patent") and 8,481,712 (the "'712 Patent"). Compl. ¶¶ 62-77, ECF 1. On August 30, 2013, Gilead filed its complaint for declaratory judgment of non-infringement and invalidity of the '499 and '712 Patents. Compl. ¶ 1, ECF 1.

8. On November 22, 2013, Merck filed its answer and amended counterclaims. Ans., ECF 62. Merck denied all allegations involving non-infringement and invalidity, *id.* at ¶¶ 66-77, and counterclaimed for a declaratory judgment of infringement of the '499 and '712 Patents, *id.* at ¶¶ 11-34.

9. On November 28, 2014, Merck filed its second amended and supplemental counterclaims. Second Am. Countercl., ECF 98. Merck repeated its previous counterclaims seeking declaratory judgment of infringement of the '499 and '712 Patents, and added additional counterclaims for infringement of the '499 and '712 Patents based on the fact that Gilead began commercially selling sofosbuvir on or about December 6, 2013. *Id.* at 1 n.1.

10. On December 15, 2014, Gilead filed its answer to Merck's second amended and supplemental counterclaims. Ans. to Second Am. Countercl., ECF 101. Gilead denied all pertinent allegations regarding infringement and invalidity, *id.* at ¶¶ 11-43, and asserted

affirmative defenses based on invalidity, laches, estoppel, waiver, and unclean hands, *id*. at 6.

11. Merck moved for summary judgment that Gilead's products (Sovaldi® and Harvoni®) that contain the active pharmaceutical ingredient "sofosbuvir" infringe the asserted claims. Merck's Mot. for SJ, ECF 167. Gilead argued that the asserted patents are invalid but conceded that if they are not invalid, then it infringes the asserted claims. Gilead's Opp. to SJ at 1, ECF 175. On February 1, 2016, the Court granted as unopposed Merck's motion for summary judgment that the sale by Gilead of Sovaldi® and Harvoni® infringes the asserted claims. Summary Judgment Order at 8, ECF 214. The Court left to a jury trial the issue of whether the asserted patents are invalid. *Id*. at 9.

12. At trial, Merck asserted claims 1 and 2 of the '499 Patent and claims 1, 2, 3, 5, 7, 9, 10 and 11 of the '712 Patent. Joint Pretrial Stmt. at 3, ECF 254.

13. From March 7-16, 2016, the Court held an eight-day jury trial on Gilead's invalidity defenses under 35 U.S.C. § 112 (lack of written description and enablement) and § 102 (derivation and prior invention). ECF 305, 306, 307, 324, 325, 327, 348, 349.

14. On March 22, 2016, the jury reached a verdict, finding the '499 and '712 Patents were not invalid. Verdict Phase 1, ECF 388. Following a three day trial on damages, ECF 386, 389, 391, the jury awarded Merck $200 million in damages for sales of Sovaldi® and Harvoni® through December 31, 2015. Verdict Phase 2, ECF 392.

15. On March 30, 2016, the Court held a bench trial on Gilead's equitable defenses. ECF 401. Prior to the bench trial, on March 22, 2016, Gilead withdrew its defenses of laches and equitable estoppel. Gilead Trial Br. at 1 n.1, ECF 368. As a result, the March 30 bench trial addressed Gilead's defenses of unclean hands and waiver. Gilead Trial Br., ECF 368; Merck Trial Br., ECF 370.

16. On April 22, 2016, Gilead filed a motion to re-open the record and allow additional evidence. ECF 410. On April 29, 2016, the Court held a hearing on Gilead's motion where the Court granted the motion and also allowed Merck to supplement the record. ECF 418.

C. Background on Hepatitis C

17. HCV was discovered in the late 1980s. Trial Tr. 191:14-17 (McHutchison).

Around 170 million people in the world and 3.2 to 3.5 million people in the United States have HCV. Trial Tr. 197:22-198:1 (McHutchison).

18. HCV is a blood borne disease. Trial Tr. at 195:19-196:16 (McHutchison). Prior to 1991, blood donations were not screened for HCV and people contracted HCV through blood transfusions. *Id*. Today, HCV is spread in other ways including the sharing of a needle or a used razor. *Id*. When a person is infected with HCV, the virus attacks and invades the liver. *Id*. Damaged liver cells are replaced with scar tissue, eventually resulting in cirrhosis and potentially causing liver cancer and requiring a liver transplant. *Id*.

19. There are seven strains, or genotypes of the HCV virus. Trial Tr. 198:2-199:2 (McHutchison). In the United States, the most common type of strain is genotype 1 (affecting between 67 and 75% of infected people) followed by genotype 2 and 3. *Id*.

20. Historically, individuals with HCV genotype 1 were treated with interferon or a combination of interferon and ribavirin. Trial Tr. 199:6-17 (McHutchison). Initially such treatment consisted of three interferon injections a week for one year and subsequently improved to one injection a week with ribavirin pills twice a day. *Id*. Side effects from this treatment resembled the flu and included fevers, chills, shakes, burning muscles, and headaches. Trial Tr. 200:6-18 (McHutchison).

21. Because of the side effects, on average, 20 percent of individuals would not participate in the treatment and 20 percent of people who started the treatment could not complete it. Trial Tr. 199:18-25; 200:19-201:1 (McHutchison). Moreover, of those who successfully completed the treatment, only about 40 percent were actually cured. *Id*.

22. In the 1990s and 2000s, significant efforts were made by various individuals and entities to find improved treatment options for HCV. *See, e.g.*, Trial Tr. 201:2-4 (McHutchison) (researched HCV treatment at Scripps Clinic and Duke University); Trial Tr. 209:15-211:13 (McHutchison) (explaining Gilead's attempts to treat HCV); Trial Tr. 254:14-255:8 (Sofia) (discussing collaboration between Roche and Pharmasset); Trial Tr. 491:19-493:1 (Otto) (explaining Pharmasset's research regarding HCV in the early 2000s); Trial Tr. 949:18-23 (Olsen) (discussing joint collaboration between Merck and Isis to research HCV treatments).

23. HCV is particularly difficult to treat for at least a few different reasons. Trial Tr. 197:4-21 (McHutchison). HCV has developed several different ways to evade the immune system and is constantly replicating. *Id.* For example, once infected, a person may have a trillion viruses in their body with half of those viruses being replaced every three to five hours. *Id.* In addition, drugs that may be effective against HCV in a laboratory setting may be unsuitable for humans due to toxic side effects. Trial Tr. 249:3-17 (Sofia). Even when a drug that is effective against HCV is discovered, it must still be delivered to the virus and liver without being converted into an inactive drug by the body. Trial Tr. 249:18-250:9 (Sofia).

D. The ’499 and ’712 Patents

24. Merck and Isis are joint assignees of the ’499 and ’712 Patents. Joint Pretrial Stmt. at 2, ECF 254.

25. The patents share a common specification, Stipulation, ECF 300; Trial Tr. 1787:20-24 (stipulation), and arose out of a joint collaboration between Merck and Isis dating from 1998-2003, Trial Tr. 961:10-17; 994:25-995:3 (Olsen). The purpose of the collaboration was to find nucleoside inhibitors of HCV RNA replication by targeting the NS5B polymerase. Trial Tr. 949:18-23 (Olsen).

26. Merck employees Dr. David Olsen, a research scientist, Trial Tr. 920:22-24 (Olsen), and Steve Carroll, an enzymologist, were some of the people that led the Merck-Isis collaboration, Trial Tr. 948:19-949:12 (Olsen).

27. As part of that years-long collaboration, the Merck-Isis scientists tested more than 2,000 nucleoside analogs, of which at least 1,000 were novel compounds made by Isis. Trial Tr. 970:21-971:2 (Olsen). The group’s work was guided in part by its analysis of structure activity relationships, which it used to identify compounds that were likely to be active. Trial Tr. 963:4-12 (Olsen). The inventors tested the compounds of the invention using an NS5B polymerase biochemical assay and a cell-based replicon assay. Trial Tr. 948:15-949:7, 969:21-970:11 (Olsen); 1561:7-15 (Wuest). The assays were performed in 96-well plates to test many compounds at one time. Trial Tr. 948:15-949:7, 1013:9-1014:1 (Olsen).

28. Philippe Durette, an in-house patent prosecutor at Merck, became involved with the

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Merck-Isis collaboration in late 2000. Trial Tr. 991:10-16 (Olsen). Dr. Durette has a bachelor's degree from Marquette University and a Ph.D. from The Ohio State University. Trial Tr. 412:14-15 (Durette). Dr. Durette did a post-doctoral fellowship for three years and afterwards started his career as a medicinal organic chemist at Merck. Trial Tr. 412:18-413:5 (Durette). After 25 years working in laboratory settings, Dr. Durette went to law school at Rutgers University and subsequently passed the bar exams in New Jersey and Pennsylvania in 1993 and 1994. Trial Tr. 413:4-13 (Durette).

29. On January 22, 2001, Dr. Durette filed U.S. Provisional Application No. 60/263,313. EX-0804. Subsequently, Dr. Durette filed additional provisional applications in April, June, and October of 2001. EX-0805, 0806, 0807.

30. The patent applications included over 150 examples depicting compounds of the invention. Trial Tr. 928:24-929:1 (Olsen).

31. On January 18, 2002, Dr. Durette filed two non-provisional patent applications having the same specification, one of which was the PCT application that led to the '499 Patent. EX-0808, 0829. These applications incorporated the provisional patent applications by reference. Trial Tr. 1587:22-1588:13 (Wuest).

32. Dr. Olsen, Dr. Carroll, Dr. Durette, and various team members were involved in drafting the 2002 patent application that eventually resulted in the '499 and '712 Patents. Trial Tr. 990:11-991:4 (Olsen).

33. On July 9, 2003, Dr. Durette filed U.S. Patent Application No. 10/258,873 (the "'499 application"), the specific application that resulted in the '499 Patent. EX-0829. It claims priority to the January 18, 2002, non-provisional patent application. EX-0001.

34. Upon initially filing the '499 application, Dr. Durette submitted a preliminary amendment presenting ten claims for prosecution. EX-0829.0247-0259. Among the ten claims for prosecution was claim 44. *Id.* Pending claim 44 covered the use of a compound from among structural formula III as defined within the claim to treat HCV. EX-0829.0257-0258. The generic structural formula III as defined in pending claim 44 was identical to a sub-embodiment of structural formula III in the specification. Compare *id. with* EX-0001.0009. That sub-

embodiment of structural formula III is limited to only single-ring, or pyrimidine, bases. *Id*. Pending claim 44 containing generic structural formula III never issued as a patent claim.

35. Between July 9, 2003 and February 7, 2005, no substantive actions took place with respect to the ’499 application. EX-8029.0001-1092. However, Dr. Durette did not forget about the ’499 application as he exchanged correspondence with the Patent Office in 2003 and 2004:

a. On October 14, 2003, Dr. Durette submitted an information disclosure statement that disclosed related applications 10/052,318 and 10/431,657. EX-8029.1070-76.

b. On December 4, 2003, the Patent Office issued a notice that that the ’499 application was missing an oath or declaration of the inventors in compliance with 37 CFR 1.497(a) and corresponding fees. EX-8029.1077-78.

c. On January 16, 2004, Dr. Durette responded to the notice by enclosing a declaration and power of attorney executed by the inventors and the appropriate fees. EX-8029.1080-88.

d. On February 11, 2004, the Patent Office issued a notice of acceptance for examination that the application complied with all the requirements of 35 U.S.C. § 371. EX-8029.1091-92.

E. The Beginning of the Pharmasset and Merck Conversations

36. During the early 2000s, Pharmasset was a research-based pharmaceutical company focused in the field of nucleoside derivatives as potential antiviral treatments, including treatments for HCV. Trial Tr. 489:21-490:3; 491:23-492:6 (Otto).

37. In 2001, Pharmasset and Merck explored potential collaboration opportunities. Trial Tr. 1019:21-1020:2 (Olsen). In order to facilitate discussions, on January 29, 2001, Pharmasset entered into a Non-Disclosure Agreement (“NDA”) with Merck. EX-2298.

38. The purpose of the NDA was to permit disclosure of “certain confidential and proprietary information concerning discovery and development of antiviral agents against flaviviruses, in particular hepatitis C virus (HCV)” for the purpose of “evaluating a possible business relationship between the Parties.” EX-2298.0002.

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39. Under the NDA, Merck agreed to hold the confidential information disclosed to it by Pharmasset in confidence and not to disclose any confidential information to any third party without the prior written authorization of Pharmasset. EX-2298.0003, ¶ 5.

40. Under the NDA, Merck agreed that it would not use Pharmasset’s confidential information for any purpose other than for evaluating a potential collaboration with Pharmasset. EX-2298.0003, ¶ 6.

41. On August 22, 2003, Pharmasset and Merck amended their NDA, again for purposes of evaluating a potential collaboration. EX-1241.0001. The August 22, 2003, Amendment stated that all terms and conditions of the January 29, 2001, Non-Disclosure Agreement would remain in full force and effect. *Id*.

42. One month later, on September 22, 2003, Pharmasset presented to Merck an overview of its HCV program. EX-2300.

43. The presentation focused on Pharmasset’s evaluation of its compound identified as PSI-6130 in both the replicon assay and the HCV NS5B polymerase assay. EX-2300.0002. PSI-6130 was first recorded by Pharmasset employee Jeremy Clark on December 6, 2002. EX-2383 at 32:11-32:17, 33:05-33:14, 34:10-34:14, 36:04-36:16, 36:24-37:12.

44. During the presentation, Pharmasset also presented to Merck data on the potency of PSI-6130 in the NS5B polymerase assay. EX-2300.0014, 0017, 0019.

45. Thus, by September 22, 2003, Merck was aware that Pharmasset’s lead compound, PSI-6130, was an NS5B polymerase inhibitor whose mechanism of action was to inhibit the NS5B polymerase enzyme.

46. On October 23, 2003, Pharmasset and Merck executed a Material Transfer Agreement (“MTA”) authorizing Merck to conduct testing and evaluation of ten Pharmasset nucleosides, including PSI-6130. EX-1231.0002, .0006. The MTA referred to the “Evaluation of Pharmasset HCV NS5B Nucleoside Inhibitor.” EX-1231.0012.

47. Under the MTA, Merck agreed to limit its use of the disclosed nucleoside compounds to testing and evaluation as set forth in the Agreement. EX-1231.0007. The MTA also barred Merck from determining the chemical structure of the nucleosides provided for testing.

Id.

48. On December 12, 2003, Pharmasset and Merck amended their MTA to include further evaluation of PSI-6130 as an HCV inhibitor. EX-1231.0003. The amendment described PSI-6130 as "a Nucleoside HCV NS5B Inhibitor" and as "the HCV NS5B polymerase inhibitor." EX-1231.0004.

49. Under the terms of these additional material transfer agreements, Merck knew that Pharmasset's PSI-6130 was an NS5B polymerase inhibitor. *Id.*

50. In January 2004, Merck tested PSI-6130 and told Pharmasset that the in vitro results were "very encouraging." EX-2302.0002. Moreover, Merck requested certain information about the structure of PSI-6130. EX-2302.0003; EX-0183.0001.

51. Maintenance of confidentiality was critically important to Pharmasset. A confidential compound's structural information is a biopharmaceutical company's "crown jewels." EX-2400 at 166:19-168:7; *see also* EX-2397 at 22:9-20.

52. Dr. Durette admitted that "[h]aving structural information is very important as to what the competition is doing in its research efforts." Durette Dep. Tr. (EX-2388) at 38:25-39:7; Trial Tr. at 359:15-18 (Durette).

53. In furtherance of the Pharmasset-Merck discussions, Merck proposed that structural information be shared with a "firewalled" Merck medicinal chemist, Dr. Wallace Ashton, to "help guide [Merck] in framing a relationship with Pharmasset in the HCV field." EX-2302.0003; EX-0183.0001.

54. In an effort to encourage Pharmasset to give Merck structural information about PSI-6130, Merck told Pharmasset that "[i]t will be very helpful to Merck if Pharmasset would consider allowing a Merck Medicinal Chemist, who is 'firewalled' from our internal HCV program, assess the lead and back-up Pharmasset compounds." EX-2302.0003.

55. A firewall is a key method to protect a confidential compound's structural information, because it limits that confidential information to only individuals not involved with the project at hand, therefore maintaining confidentiality. EX-2400 at 166:19-168:7.

56. Merck understood that the purpose of the firewall was to protect Pharmasset's

confidential structural information about its lead compound, PSI-6130. EX-2302.0003; *see also* EX-2397 at 24:08-24:11, 24:14-16.

57. Pharmasset only agreed to provide more information about the structure of PSI-6130 to Merck personnel who were within the firewall (i.e., “firewalled”). EX-2302.0001-.0002.

58. A firewalled person would not have any involvement with Merck’s internal HCV program. EX-2302.0001.

59. Thus, Pharmasset was willing to provide structural information about PSI-6130 to Merck because there was a confidentiality agreement in place between the parties and the information would be firewalled. EX-2302.0001.

60. On February 4, 2004, Pharmasset provided information to firewalled Merck chemist, Dr. Wallace Ashton, disclosing that PSI-6130 was a cytosine base containing nucleoside, without a N=O bond, and with a 5’ hydroxyl group. EX-0046.001; EX-0047.0001-2.

61. In communicating that structural information, Pharmasset reminded Dr. Ashton that the information was only being shared with him because he was firewalled. EX-0047.0001.

62. Dr. Ashton understood that, as a firewalled chemist receiving structural information about PSI-6130, he was not permitted to communicate specifics of the compound’s structure to anyone outside the firewall. EX-2397 at 24:8-26:4, 34:8-12.

63. Despite the NDA, MTA and firewall restrictions, in March 2004, Merck directed Dr. Durette, one of its in-house patent attorneys, to participate in a due diligence call with Pharmasset. Trial Tr. at 355:22-360:15 (Durette); EX-0153.

64. As discussed *supra* Findings of Fact (“FOF”) ¶¶ 28-29, since 2001, Dr. Durette had been the attorney responsible for prosecuting patent applications related to nucleoside analogs for the treatment of HCV based on the Merck-Isis HCV collaboration, including the ’499 application. Trial Tr. at 328:21-24 (Durette). These patent applications disclosed NS5B polymerase inhibitors. EX-0001; EX-0808.

65. On March 11, 2004, one month after the Patent Office issued the ’499 application’s notice of acceptance for examination, Dr. Durette was copied on an e-mail from Pamela Demain, a Merck corporate licensing specialist, regarding the upcoming March 17, 2004, due diligence call

with Pharmasset. Trial Tr. 356:20-357:10 (Durette). The other recipients of this e-mail were Mervyn Turner, Anthony Ford-Hutchinson, Barbara Yanni, Malcolm Maccoss, Daria Hazuda, David Olsen, Scott Kauffman, Doug Pon, Frank Potter, Michael Rabinowitz, Durga Bobba, and Linda Stefany. The e-mail evidences Merck's intention that Dr. Durette would participate in the due diligence call.

66. In that March 11, 2004, e-mail, Ms. Demain noted that "Pharmasset has not yet permitted us to review the structure of PSI-6130." EX-0153.0001.

67. In that March 11, 2004, e-mail, Ms. Demain wrote "[a]s a first step, Phil Durette will view the structure during a patent due diligence meeting on March 17[, 2004]." EX-0153.0001.

68. Ms. Demain's March 11, 2004, e-mail attached a proposed Merck-Pharmasset term sheet. She stated in the e-mail that the term sheet had been reviewed by Dr. Durette. Trial Tr. at 2499:1-2500:1 (Demain); EX-0153.0001.

69. The proposed term sheet that Dr. Durette reviewed stated that Pharmasset's "lead compound PSI 6130…is a chain terminator of HCV polymerase." EX- 2394.0002; Trial Tr. at 2500:5-21 (Demain).

70. A chain terminator of HCV polymerase is the same type of compound for which Dr. Durette was prosecuting patent applications for Merck, and the same type of compounds which were the subject of the Merck-Isis collaboration. Trial Tr. at 951:12-955:21 (Olsen) (describing collaboration as focused on chain terminators).

71. From his review of the term sheet and Ms. Demain's email, Dr. Durette knew, before the March 17, 2004, patent due diligence phone call with Pharmasset, that:

a. PSI-6130 was Pharmasset's lead compound, EX-0153.0001; EX-2394.0002; Trial Tr. at 1430:9-18 (Demain);

b. Pharmasset believed PSI-6130's value was "in excess of $100 million total," EX-153.0001;

c. he would learn the structure of PSI-6130 during the March 17, 2004 phone call, EX-0153.0001;

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d. PSI-6130 was a chain terminator of the HCV polymerase, Trial Tr. at 2500:17-2501:4 (Demain); EX-2394.0002; and

e. PSI-6130 was an NS5B polymerase inhibitor, Trial Tr. at 2500:17-2501:4 (Demain); EX-2394.0002.

72. In light of the facts recited *supra* FOF ¶¶ 64-70, the Court finds that Dr. Durette knew, before the March 17, 2004, phone call, that any information he learned about Pharmasset's PSI-6130 nucleoside analog compound would overlap with the subject matter of his patent prosecution docket for Merck, thereby creating a conflict. Trial Tr. at 354:14-355:16; 364:11-365:11, 375:7-23 (Durette).

73. Furthermore, Dr. Durette did not qualify as a firewalled individual; he was prosecuting patents from the Merck-Isis collaboration. *See, e.g.*, Trial Tr. 990:11-991:4 (Olsen).

74. Merck's corporate policy forbids Merck's patent prosecutors from participating in licensing discussions in an area related to their prosecution work. Durette Dep. Tr. (EX-2388) at 38:25-39:7.

75. Dr. Durette knew, before the March 17, 2004, due diligence phone call with Pharmasset, that learning the structure of PSI-6130 would overlap with his responsibilities in prosecuting patent applications concerning the Merck-Isis collaboration, including the '499 application and violate corporate policy.

76. Thus, in light of the facts recited *supra* FOF ¶¶ 64-75, the Court finds that it was improper for Merck to plan to have its employee Dr. Durette participate on the March 17, 2004, due diligence call with Pharmasset.

F. The Phone Call

77. On March 17, 2004, a due diligence phone call was held between Merck and Pharmasset. EX-2098.

78. The Merck participants on the March 17, 2004, phone call were Dr. Durette and Dr. Pon. *Id.* The Pharmasset participants on the March 17, 2004, phone call were Alan Roemer, Dr. Raymond Schinazi, and Bryce Roberts. *Id.*

79. This March 17, 2004, phone call occurred barely one month after Dr. Durette

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received the ’499 application’s notice of acceptance for examination. Trial Tr. 354:24-355:16 (Durette).

80. Mr. Roemer took notes during the call. EX-2098.

81. During the March 17, 2004, call, Dr. Durette learned the structure of PSI-6130. Trial Tr. at 431:7-14 (Roemer); Trial Tr. at 347:9-22 (Durette); EX-2098.

82. At the beginning of the call, Dr. Schinazi reminded everyone that it was a firewalled conversation. Trial. Tr. at 382:8-12 (Durette); EX-2098.0001 (RFS: “Firewall”). This meant that no one from Merck on the telephone call should have been involved in Merck’s HCV program. EX-2302.0003.

83. Before Pharmasset revealed the structure of PSI-6130, Dr. Durette did not tell Pharmasset that he was prosecuting patents in the same field of HCV nucleoside analogs. Trial Tr. at 435:7-12 (Roemer); EX-2098; Trial. Tr. at 382:8-383:6 (Durette).

84. Merck violated its own company policy by directing Dr. Durette to participate in the due diligence phone call with Pharmasset. Durette Dep. Tr. (EX-2388) at 38:25-39:7.

85. Mr. Roemer’s notes reflect that after initial information about the structure of PSI-6130 was disclosed, Dr. Durette stated that the information he learned “seems quite related to things that I’m involved with,” and that he “need[ed] to have a conversation with his supervisor.” EX-2098.0002. Moreover, according to Mr. Roemer’s notes, Dr. Durette clarified that he was “personally conflicted; not the company.” EX-2098.

86. At the end of the call, Mr. Roemer again reminded the Merck attendees that this was a firewalled conversation, and sought confirmation that Dr. Durette and Dr. Pon were within the “firewall” of the Confidentiality Agreement. Trial Tr. 382:8-18 (Durette); Trial Tr. at 434:1-24 (Roemer); EX-2098.0002.

87. At the end of the call, both Dr. Durette and Dr. Pon specifically stated that each of them was within the firewall. Trial Tr. at 434:1-20 (Roemer); EX-2098.0002.

88. After the March 17, 2004, call, neither Merck nor Dr. Durette ever informed Pharmasset that Dr. Durette was not in fact firewalled and was in fact prosecuting Merck’s patents in the same field.

89. At his deposition, Dr. Durette testified that if he had learned the structure of PSI-6130, then according to Merck's procedures and policies, he would have had to turn his prosecution of Merck's HCV patents over to another attorney. Durette Dep. Tr. at 201:23-202:16, ECF 410-3.

90. Instead of withdrawing from prosecution, Dr. Durette continued to prosecute Merck's HCV patent applications and write new claims that targeted Pharmasset's work. The new claims that targeted Pharmasset's work were based on the information he learned on the March 17, 2004, patent due diligence call.

91. The Court finds that:

a. Dr. Durette's statements to Pharmasset on the March 17, 2004, call about being within the firewall were untrue;

b. Merck, through Dr. Durette and Dr. Pon, knowingly misrepresented to Pharmasset that Dr. Durette was firewalled;

c. it was a violation of the Merck-Pharmasset firewall for Dr. Durette to participate on the March 17, 2004, call;

d. it was improper for Merck and Dr. Durette never to have informed Pharmasset that Dr. Durette was not within the firewall and was in fact prosecuting Merck's patents in the same field;

e. after Dr. Durette learned the structure of PSI-6130 on the March 17, 2004, phone call, Merck was required to recuse Dr. Durette from any further prosecution of the Merck-Isis patent applications, in order to comply with Merck's obligations under the NDA, EX-2298, EX-0124, and the firewall; and

f. Merck and Dr. Durette's failure to recuse Dr. Durette from further prosecution of the Merck-Isis patent applications was an improper business practice.

92. Neither Merck nor Dr. Durette has provided any explanation for why Dr. Durette was not excluded from further prosecution of the Merck-Isis patent applications after learning the structure of PSI-6130 during the firewalled patent due diligence call.

G. Dr. Durette's Continued Prosecution of the '499 and '712 Patents

93. On the March 17, 2004, patent due diligence call, Dr. Durette was told by Pharmasset that Pharmasset's patent application would be publishing in November 2004. EX-2098.0002.

94. Pharmasset's patent application naming Jeremy Clark as the inventor and disclosing the structure of PSI-6130 published on January 13, 2005. EX-0155.

95. As of February 1, 2005, the Patent Office had not allowed the then-pending claims of the '499 application. EX-0829.

96. On February 1, 2005, Dr. Durette cancelled all then-pending claims of the '499 application and submitted the two new, narrower claims (53 and 54) for prosecution. EX-0156.0004.

97. None of the listed inventors on the '499 Patent was involved in Dr. Durette's patent claiming strategy or the change in claims that took place on February 1, 2005. Bhat Dep. Tr. (EX-2377) at 100:11-17; Eldrup Dep. Tr. (EX-2378) at 55:24-56:6; Carroll Dep. Tr. (EX-2379) at 129:1-10; Cook Dep. Tr. (EX-2376) at 255:11-15; Olsen Dep. Tr. (EX-2380) at 213:18-21. This is despite the fact that several Merck-Isis team members had been involved with drafting the initial application. Trial Tr. 990:11-991:4 (Olsen) (explaining Dr. Olsen, Dr. Carroll, Dr. Durette, and various team members were involved in drafting the 2002 patent application that eventually resulted in the '499 and '712 Patents).

98. The then-pending claims had not been rejected by the patent examiner at the Patent Office, and the examiner had not asked Dr. Durette to narrow the claims. *See* EX-8029. Dr. Durette did that on his own. Trial Tr. at 372:18-23 (Durette).

99. The two new, narrower claims Dr. Durette submitted on February 1, 2005, do not cover any compound tested by Merck and Isis during the Merck-Isis collaboration. Stipulation, ECF 300; Trial Tr. 554:6-10 (stipulation).

100. The two narrowed claims issued as claims 1 and 2 of the '499 Patent. EX-0156.0004; *see also* EX-0001.0071.

101. Dr. Durette waited until Pharmasset published the structure of PSI-6130 and then

wrote claims to cover Pharmasset's invention. Trial Tr. at 369:24-374:4, 389:25-390:14; 417:1-19 (Durette).

102. The Court finds that Dr. Durette waited to amend the claims in the '499 Patent until Clark application was published to give the appearance that he learned it from a public source.

103. Dr. Durette has admitted that he would not have been able to associate any structure in the Pharmasset application as the structure of PSI-6130 unless he knew the structure of PSI-6130 beforehand. Durette Dep. Tr. at 53:1-6, 53:22-54:5, ECF 410-3.

104. The Court finds that Dr. Durette would not have written new claims to cover PSI-6130 in February 2005 but for his improper participation on the March 17, 2004 patent due diligence call and learning the structure of PSI-6130 ahead of the structure being published.

105. Additionally, in further violation of Merck's corporate policy and the Merck-Pharmasset firewall, it was improper for Merck to allow Dr. Durette to prosecute the '712 Patent after having participated on the March 17, 2004, call and learning the structure of PSI-6130. Dr. Durette filed the application that resulted in the '712 Patent in February 2007. EX-2375 (Bergman Dep. Tr.) at 26:16-24, 27:03-06; EX-0192.0003.

106. The '499 and '712 Patents share a common specification. Stipulation, ECF 300; Trial Tr. 1787:20-24 (stipulation).

H. Dr. Durette's Deposition

107. Dr. Durette was deposed in this case on May 8, 2015. Durette Dep. Tr. at 1, ECF 410-3.

108. Dr. Durette was Merck's designated Fed. R. Civ. P. 30(b)(6) corporate representative on issues related to the preparation and prosecution of the patent application leading to the '499 patent-in-suit, including all reasons for amending any pending claim during prosecution. Durette Dep. Tr. at 181:25-182:16, ECF 410-3.

109. At the deposition, Dr. Durette was represented by Merck's outside counsel. Durette Dep. Tr. at 7:16-19, ECF 410-3.

110. Leading up to his deposition, Dr. Durette met with Merck's outside and inside counsel for two full days of preparation, six to seven hours for each day. Durette Dep. Tr. at

10:19-11:11, ECF 410-3.

111. Dr. Durette spent an additional 8-10 hours on his own preparing for the deposition. *Id*.

112. Dr. Durette testified at his deposition that he had the same memory of events before and after looking at documents related to the Merck HCV program. Durette Dep. Tr. at 14:8-15:11, ECF 410-3.

113. During the deposition, Dr. Durette was questioned about his participation in the March 17, 2004, patent due diligence call. Durette Dep. Tr. (EX-2388) at 30:21:31:10.

114. When asked about the March 17, 2004, call at the deposition, Dr. Durette denied ever having been on such a call. When asked whether he was sure that he was not on the March 17, 2004, call, Dr. Durette unequivocally answered yes.

> Q: …In March of 2004 were you involved in any discussion with Pharmasset whereby you were told what the structure was for their 6130 compound?
>
> A: No.
>
> Q: You're sure of that?
>
> A: Yes.

Durette Dep. Tr. (EX-2388) at 30:21-31:3.

115. Dr. Durette also stated that he was "positive" that the structure of PSI- 6130 was "never" revealed to him:

> Q: How are you so sure 11 years later that you were never told what the structure was for the 6130 compound?
>
> A: The structure was not revealed to me by individuals at Merck or otherwise. I'm positive of that. I never saw a structure of the Pharmasset compounds until it published later on in time.

Durette Dep. Tr. (EX-2388) at 31:4-31:10.

116. Dr. Durette did not say that he did not remember a call or that he could not be sure, but definitively stated that he was sure he was never on the call and "positive" that he never saw

the structure of PSI-6130 prior to it being published later. *Id.*

117. Later in the deposition, Dr. Durette also definitively stated that "I never participated in a due diligence meeting on March 17 because the due diligence component of this potential deal was assigned to another attorney, so there was—I did not participate in any meeting of due diligence on March 17." Durette Dep. Tr. (EX-2388) at 37:13-18.

118. Dr. Durette offered several reasons why he never learned the structure of PSI-6130 in March 2004.

> Q: How can you be so sure of that memory?
>
> A: Because I was not part of the patent due diligence for the structure, so I would not have been privy to any revelation of the structure to me as a patent attorney working on a related docket. So this was assigned to another person. So I would not have participated in a phone call wherein it was a potential for the revelation of the structure to Merck counsel.
>
> Q: Why would that have been inappropriate for you to have been told the structure of 6130?
>
> A: Because I was prosecuting a docket which had potential a conflict with Pharmasset's IP positions on the subject matter.

Durette Dep. Tr. (EX-2388) at 38:1-38:13.

119. Dr. Durette acknowledged at his deposition that it was against Merck's company policy to have a Merck patent prosecutor participate in licensing discussions in a related area. Durette Dep. Tr. (EX-2388) at 38:25-39:07.

120. Dr. Durette explained at his deposition "[h]aving structural information is very important as to what the competition is doing in its research efforts. We had a policy at Merck on a particular docket area if there were potential licensing opportunities in a related area, that due diligence would be assigned to a non – an attorney that was not prosecuting a particular docket in a related area." Durette Dep. Tr. (EX-2388) at 38:25-39:7.

121. Dr. Durette acknowledged at the deposition that learning the structure of PSI-6130 would "have tainted [his] judgment as to what claims to pursue in the Merck/Isis collaboration."

Durette Dep. Tr. (EX-2388) at 38:21-38:24.

122. Pharmasset's patent application, known as the Clark application, published on January 13, 2005. EX-0155. When Pharmasset's patent application published on January 13, 2005, it disclosed a "large collection of compounds." Durette Dep. Tr. at 52:25, ECF 419-1. In Dr. Durette's words, PSI-6130 was but one structure among a "plethora of compounds" disclosed in the patent application. Durette Dep. Tr. at 53:25-54:1, ECF 419-1.

123. Without knowing the structure of PSI-6130 in advance of the application, Dr. Durette would not have been able to associate any compound in the patent application published on January 13, 2005, with PSI-6130. Durette Dep. Tr. at 52:19-23, ECF 419-1.

> Q: How is it that you know that you would not in January of 2005 have realized that Paragraph 0168, that chemical structure there, was 6130?
>
> A: Because this was one compound out of a plethora of compounds in the publication.
>
> Q: Now, if you had been told prior to this publication what the structure of 6130 was, then you would have been able to match it up, right?
>
> A: Yes.

Durette Dep. Tr. at 53:25-54:5, ECF 410-3.

124. Having denied being on the March 17, 2004, due diligence call, Dr. Durette was shown Ms. Demain's March 11, 2004 e-mail which said that he was specifically chosen by Merck to receive the structure of PSI-6130 on a March 17, 2004, patent due diligence call. Durette Dep. Tr. (EX-2388) at 37:02-18; EX-0153. He was asked if this refreshed his recollection. Durette Dep. Tr. (EX-2388) at 37:02-18.

125. In the face of Ms. Demain's e-mail, Dr. Durette still denied being on the call, contending "[t]hat was Pamela's evaluation of the time, but I never participated in a due diligence meeting on March 17 because the due diligence component of this potential deal was assigned to another attorney, so there was – I did not participate in any meeting of due diligence on March 17." Durette Dep. Tr. (EX-2388) at 37:13-18.

126. Dr. Durette was then shown a May 20, 2004, letter and asked if that letter refreshed

his recollection about the March 17, 2004, call. Durette Dep. Tr. at 168:5-16, ECF 410-3. The May 20, 2004, letter contained a list of things Pharmasset wanted returned, including "notes from a March 17, 2004, telephone conference regarding PSI-6130 patent due diligence with [Doug Pon] and Phil Durette." *Id*.

127. Dr. Durette still denied being on the call, stating that it was his sworn testimony that he was not made aware of the structure of PSI-6130 on the March 17, 2004, call, and that he remembered that clearly. Durette Dep. Tr. at 168:24-169:18, ECF 410-3.

128. At the time of his deposition, no one told Dr. Durette that Pharmasset's Alan Roemer had taken contemporaneous notes of that March 17, 2004, patent due diligence phone call. Trial Tr. at 380:22-25 (Durette).

129. Mr. Roemer was deposed by Merck's counsel on May 24, 2015. Roemer Dep. Tr. at 1.

130. At Mr. Roemer's deposition, his notes were used as an exhibit, and Gilead's counsel asked Mr. Roemer about the call that occurred on March 17, 2004. Mr. Roemer testified that Dr. Durette participated in the call and that Dr. Durette was provided the structure of PSI-6130 on that call. Roemer Dep. Tr. at 233:3-22.

131. Between May 24, 2015, the date of Mr. Roemer's deposition, and March 8, 2016, the start of trial, Merck never indicated that Dr. Durette's deposition testimony was untruthful or incorrect.

132. In his opening statement at trial, on March 8, 2016, Merck's counsel stated that Merck would not dispute that Dr. Durette was on the March 17, 2004, call with Pharmasset. Trial Tr. at 178:5-179:1 (Merck's opening statement). Counsel for Merck further told the jury that Dr. Durette did not know that the compound that Pharmasset was going to disclose was within the scope of what Merck was working on. Trial Tr. 178:8-11 (Merck's opening statement). That representation of Dr. Durette's pre-call knowledge was incorrect. *See infra*, FOF ¶¶ 142-143.

133. Gilead first learned of Dr. Durette's new story during Dr. Durette's examination at trial.

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I. Dr. Durette's Trial Testimony

134. Dr. Durette was outside the subpoena power of this Court and Gilead could not force his attendance at trial. Final Pretrial Conf. Tr. at 42:5-17, ECF 280. Merck, knowing about Dr. Durette's deposition testimony, voluntarily brought Dr. Durette to trial to testify on its behalf.

135. At trial, Dr. Durette provided key testimony for Merck on validity issues, including written description of the '499 Patent. Trial Tr. 391:10-404:19 (Durette). For example, Dr. Durette testified that his amendment to the '499 Patent "was fully supported by the specification," Trial Tr. 403:15-17 (Durette), and that "[Merck] had support for written -- written description support in terms of how to make the[structure] and how to use them." Trial Tr. 410:11-15 (Durette).

136. At trial, Dr. Durette said that his memory of the March 17, 2004, patent due diligence call became refreshed in January 2016 when he reviewed the deposition exhibits in preparation for trial. Trial Tr. at 386:6-15 (Durette).

137. When confronted with his deposition testimony that he had not participated in the Pharmasset-Merck due diligence call, Dr. Durette said he was relying too much on his memory. Trial Tr. at 344:8-17 (Durette).

138. Dr. Durette attempted to explain away his deposition testimony by stating that he had a lapse in memory and "over concluded" based on his memory. Trial Tr. at 344:18-345:7, 347:9-348:1 (Durette).

139. When asked about the March 17, 2004, call at trial, Dr. Durette said that the answers he gave at the deposition were "based on my lack of recollection of the events and I over concluded that I had – that I had not seen the structure." Trial Tr. at 344:1-345:7, 347:9-22 (Durette).

140. Dr. Durette further testified at trial that Pamela Demain, Merck's director of corporate licensing, asked him to attend the March 17, 2004, call. Trial Tr. at 355:17:23, 375:12-19 (Durette).

141. Ms. Demain credibly testified that she did not ask Dr. Durette to attend the call. Trial Tr. at 1404:14-1405:8 (Demain). Instead, Ms. Demain explained she was simply acting as a

messenger when she sent her March 11, 2004, e-mail and she did not know who asked Dr. Durette to be on that call. Trial Tr. at 1405:1-8 (Demain). The Court concludes that Dr. Durette's testimony was not credible on this point.

142. Dr. Durette also asserted at trial that before the due diligence call, while he knew PSI-6130 was a nucleoside, he did not know that PSI-6130 was an inhibitor of the NS5B polymerase. Trial Tr. at 364:13-18, 365:13-21, 367:13-368:6 (Durette).

143. Contrary to that testimony, Ms. Demain credibly testified that Merck and Dr. Durette did know that PSI-6130 was a nucleoside NS5B polymerase inhibitor. Trial Tr. at 2498:2-4, 2499:1-2501:4 (Demain); EX-0153; EX-2394. The Court concludes that Dr. Durette's testimony was not credible on this point.

144. Dr. Durette stated at trial that he went into the March 17, 2004, call knowing that he would receive the structure of PSI-6130 but he "did not think it was going to be likely that it would be on the subject matter that was related to the – my HCV docket." Trial Tr. at 350:25-351:9 (Durette).

145. Contrary to that testimony, Dr. Durette was prosecuting patents directed to nucleoside NS5B polymerase inhibitors, Trial Tr. at 367:13-23 (Durette), and he knew going into the call that PSI-6130 was a nucleoside NS5B polymerase inhibitor. EX-0001.0001; EX-0808; EX-2394.0002; Trial Tr. at 2498:2-4, 2499:1-2501:4 (Demain). Again, the Court concludes that Dr. Durette's testimony was not credible on this point.

146. At trial, Dr. Durette for the first time said that he had had a pre-call meeting with his manager and they had determined that it was fine for him to learn the structure of PSI-6130 because Dr. Durette was prosecuting patents related to nucleosides with a certain mechanism of action, NS5B polymerase inhibitors. Trial Tr. at 360:16-361:21 (Durette); *see also* Trial Tr. at 365:13-21, 367:13-368:14 (Durette). Specifically, Dr. Durette testified that his manager and he decided it was fine for Dr. Durette to learn the structure of PSI-6130 for several reasons: (1) HCV has "many different target enzymes"; (2) nucleosides for HCV is a "very broad area"; (3) nucleosides that attack different enzymes can have "totally different structures" and different "structure types" with "different overall mechanisms of action." *Id*. Dr. Durette offered no

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explanation for this sudden clear memory.

147. Contrary to that testimony, Merck, and Dr. Durette in particular, knew before the meeting that PSI-6130 was a nucleoside NS5B inhibitor with the same mechanism of action of the compounds for which he was seeking patent protection on behalf of Merck and Isis. EX-2300; EX-1231; EX-0153; EX-2394; EX-0090; Trial Tr. at 2498:2-4, 2500:5-2501:4 (Demain). Ms. Demain credibly testified that Dr. Durette knew this fact. Trial Tr. at 2500:5-2501:4 (Demain). The term sheet attached to the e-mail from Ms. Demain, which Dr. Durette reviewed, states that: "Until then, this amount [of the proposed license] is based on the following assumptions: . . . That lead compound PSI-6130 . . . is a chain terminator of HCV polymerase . . . ." EX-2394.0002. The Court concludes that Dr. Durette's testimony was not credible on this point.

J. Clark Publication

148. Pharmasset's patent application, known as the Clark application, published on January 13, 2005. EX-0155.

149. When Pharmasset's patent application published on January 13, 2005, PSI-6130 was but one structure among a number of structures disclosed in the patent application. EX- 0155.

150. At trial, Dr. Durette said that seeing the Clark application in 2005 caused him to think that any confidentiality obligations he had under the NDA had terminated. Trial Tr. at 369:24-370:14 (Durette).

151. Contrary to that testimony, at his deposition, Dr. Durette testified that he had no memory of when he saw Pharmasset's published patent application, and that in any event, he never associated that application with the structure of PSI-6130. Durette Dep. Tr. at 48:15-20, 51:25-52:1, ECF 410-3.

152. In fact, at his deposition, Dr. Durette—who was Merck's corporate representative with respect to the February 1, 2005 claim amendment—testified that he was not sure if he saw the Clark publication before the February 1, 2005 claim amendment:

> Q: You're just not sure if you saw the Clark publication before February 1, 2005?
>
> A: Correct.

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Durette Dep. Tr. at 67:22-24, ECF 410-3; *see also id.* at 65:14-67:24, ECF 410-3.

153. At trial, Dr. Durette said that seeing the Pharmasset patent application must have been a triggering event that led him to reexamine his docket and look at the '499 Patent application. Trial Tr. at 390:23-391:9 (Durette).

154. Contrary to that testimony, at his deposition, Dr. Durette further testified that Pharmasset's application would have had no impact, even if he had seen the application, on his amendment of Merck's claims. Durette Dep. Tr. at 71:11-72:3.12, ECF 410-3.

155. Dr. Durette also testified at his deposition that he would not have realized that the structure disclosed in paragraph 0168 of the Pharmasset application was PSI-6130 because it was just "one compound out of a plethora of compounds." Durette Dep. Tr. at 53:22-54:5, ECF 410-3.

156. Dr. Durette further testified at his deposition that he never associated the published Clark chemical structure with PSI-6130. Durette Dep. Tr. at 52:19-23, 53:1-6, ECF 419-1.

157. Dr. Durette acknowledged at his deposition that if had he been told the structure of PSI-6130 prior to the patent publication, then he would have been able to match up PSI-6130 to the structure disclosed at paragraph 0168. Durette Dep. Tr. at 54:2-5, ECF 410-3. However, at his deposition, Dr. Durette testified he was not sure he even saw the Clerk publication before February 1, 2005. Durette Dep. Tr. at 65:14-67:24, ECF 410-3.

K. Amendment of the Claims

158. Dr. Durette canceled all pending claims in the '499 Patent application in February 2005 and drafted two new claims to cover PSI-6130. Trial Tr. 375:24-376:10 (Durette). The Court finds that he did so because he had learned the structure of PSI-6130 on the March 17, 2004, call.

159. At deposition, Dr. Durette testified that he was not sure he saw the Clark publication prior to amending the claims. Durette Dep. Tr. 48:10-52:1, ECF 410-3. Given the timing of his amendment, mere days after the Clark publication, and his contradictory and evasive testimony at trial, the Court finds Dr. Durette's deposition testimony is not credible.

160. At his deposition and on cross examination at trial, Dr. Durette insisted that he filed the two, narrower claims in the '499 application simply to "expedite" prosecution. Trial Tr. at

374:7-375:2 (Durette).

161. At trial, on direct examination by Merck’s counsel, Dr. Durette stated that he amended the ’499 claims to focus on “get[ting] allowance on the subject matter that was most important to the [Merck-Isis] collaboration.” Trial Tr. at 404:14-19 (Durette).

162. Dr. Durette’s changing and evasive explanations for why he narrowed the claims undermine his testimony. The Court finds his testimony to be not credible.

163. Additionally, Dr. Durette’s claim that he amended the ’499 claims to focus on “get[ting] allowance on the subject matter that was most important to the [Merck-Isis] collaboration” is contrary to the evidence and is not credible because Merck never tested any of the claimed compounds. Stipulation, ECF 300; Trial Tr. 554:6-10 (stipulation).

164. Neither Merck nor Isis tested a single compound falling within the new claims of the ’499 Patent during the Merck-Isis collaboration that ended in 2003. Stipulation, ECF 300; Trial Tr. 554:6-10 (stipulation).

165. Merck did not test a single compound claimed in the ’499 Patent until August 2005, after Jeremy Clark’s patent application published, and after Dr. Durette added the two new claims to the ’499 Patent. Trial Tr. at 576:1-22 (Seeger); Stipulation, ECF 300; Trial Tr. 554:6-10 (stipulation).

166. Neither Merck nor Isis made a 2’-methyl up, 2’-fluoro down pyrimidine or purine nucleoside compound, tested such a compound, or used such a compound during the Merck-Isis collaboration that ended in 2003. Bennett Dep. Tr. (EX-2381) at 123:15-124:01, 124:06-21; Duffy Dep. Tr. (EX-2382) at 46:22-25; Trial Tr. at 576:1-22 (Seeger); Stipulation, ECF 300; Trial Tr. 554:6-10 (stipulation).

167. The Court finds that it is not credible that compounds that were never made, used, or tested during a collaboration were considered by Merck to be the most important work of the collaboration.

168. The only 2’-methyl up, 2’-fluoro down compound proposed by Merck and Isis was never made, does not fall within the claims of the ’499 Patent, and was a “lower priority.” Song Dep. Tr. (EX-2385) at 175:16-21, 177:1-5, 178:4-9, 189:10-18; *see also* Trial Tr. at 736:8- 17

(Secrist); Trial Tr. at 982:9-17, 983:9-984:20 (Olsen); EX-0036.0056; EX-1543.0003; Bennet Dep. Tr. (EX-2381) at 111:2-10, 123:9-12, 124:6-9.

169. Merck did not make a 2’-methyl up, 2’-fluoro down purine or pyrimidine compound until August 2005, seven months after Mr. Clark’s patent application published, and six months after Dr. Durette filed new patent claims to cover such compounds in February 2005. Trial Tr. at 1130:12-17; Duffy Dep. Tr. (EX-2382) at 46:22-25.

170. The Court finds Dr. Durette’s testimony that the two new, narrower claims he wrote in the ’499 Patent were to protect Merck’s “most important work” is not credible and is false.[2]

L. The ’712 Patent

171. The ’712 Patent was filed on February 2, 2007 as U.S. Patent Application No. 11/701,682 (the “’712 application”) by Dr. Durette. EX-0002.0001; EX-0192.0003; Bergman Dep. Tr. (EX-2375) at 25:5-27:6.

172. While Mr. Jeffrey Bergman, Merck’s in-house patent attorney, took over prosecution of the ’712 Patent application in 2011, Dr. Durette was involved in prosecuting the application prior to that. Bergman Dep. Tr. (EX-2398.0001) at 17:1-7, 17:25-18:7.

173. Merck asserted both the ’499 and ’712 Patents in this action and Dr. Durette was Merck’s 30(b)(6) witness on the prosecution of the ’499 Patent, which shares the same specification as the ’799 Patent. Durette Dep. Tr. 181:25-182:16, ECF 410-3.

M. Waiver

174. Merck and Pharmasset had discussions in 2003-2004 about the possibility of Merck in-licensing Pharmasset’s lead compound PSI-6130. Trial Tr. 1402:6-24 (Demain). Merck scientists were interested in PSI-6130 because they believed that combination therapy was the future of HCV treatment and that PSI-6130, if successful, might be used with Merck’s own MK-

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[2] Although Gilead introduced evidence of Dr. Durette’s work on a related patent application, the ’224 Patent application, the Court did not consider it in assessing Merck’s misconduct. There are various legitimate reasons why a patentee may choose to abandon a pending application and the fact that Merck and Dr. Durette chose to abandon the prosecution of the ’224 Patent application is not relevant.

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0608 compound and other anti-HCV drugs. Trial Tr. 1056:25-1058:2 (Olsen).

175. There is no evidence that Merck communicated to Pharmasset that Merck was waiving its patent rights during the 2004 timeframe. And no one from Pharmasset ever communicated to Merck that it believed Merck waived its patent rights. Trial Tr. 2482:2-18 (Demain). Nothing Merck did could be construed as a waiver of patent rights in 2004.

176. Beginning in 2008 through 2011, there were several years of on-again, off-again negotiations between Merck and Pharmasset over partnering opportunities in the antiviral space including in the HIV, Hepatitis B, and Hepatitis C areas. Trial Tr. 1405:16-1406:11 (Demain). On numerous occasions, Pharmasset contacted Merck to see if Merck was interested in a deal. *Id.*; *see also* Trial Tr. 1407:5-1408:15 (Demain); EX-1675 (timeline of Merck-Pharmasset discussions).

177. In the 2008 period, the driver of discussions was Pharmasset's Hepatitis B drug Clevudine in late-stage clinical studies. Trial Tr. 1405:16-1406:11 (Demain). In October 2008, Merck offered to license Clevudine along with Pharmasset's anti-HCV program, or alternatively, to purchase Pharmasset for $625 million. EX-1768; EX-0093 at 1-2. In its letter, Merck pointed out that one advantage of Merck acquiring Pharmasset would be that Pharmasset would get "[t]he ability to leverage Merck's intellectual property estate to reduce uncertainty and enhance the value of the Pharmasset assets going forward." EX-1768 at 2; EX-0093 at 2. Merck conveyed to Pharmasset that Pharmasset would benefit by no longer having to concern itself with the risk associated with Merck's blocking patents. Trial Tr. 1409:17-1411:1 (Demain); Trial Tr. 2483:16-2484:19 (Demain).

178. Ms. Demain testified without contradiction that Merck's patents were always in the background of the discussions with Pharmasset. Trial Tr. 2482:2-11 (Demain). Ms. Demain dealt primarily with Pharmasset's head of licensing, Abel De la Rosa. Trial Tr. 2482:19-21 (Demain). The two discussed Merck's patents generally, but there was no ambiguity that one of the patents at issue was the '499 Patent series. Trial Tr. 2482:22-2483:2; 2520:21-2521:14 (Demain) (explaining that "there's no ambiguity" about which patents were discussed with Dr. De la Rosa "because there were two patents, and it was very clear what we were speaking about"). No Pharmasset witness testified to having any other understanding of these discussions. Ms. Demain conveyed to

Pharmasset that there was unique value in Pharmasset partnering with Merck because Pharmasset would gain access to Merck's patents. Trial Tr. 2521:2-8 (Demain).

179. The documents corroborate Ms. Demain's account. On October 8, 2009, in an internal memorandum, Pharmasset stated that "[a]ll things considered, Merck is the ideal strategic partner for PSI-7851 [sofosbuvir] and Pharmasset. Consolidating nucleos(t)ide IP would lower the legal risk of this program." EX-1770 at 2 (emphasis added), App'x at 35.

180. Beginning around October 2009, and carrying through to August 2010, Pharmasset and Merck exchanged draft term sheets that would make Merck a development and marketing partner of sofosbuvir for which Merck would pay Pharmasset, and in which Pharmasset would get a cross-license to Merck's patents. EX-1622 (October 2009); EX-1625 (December 2009 draft); EX-1630 (April 2010 draft); EX-2390 (July 2010 draft); EX-1652 (referencing forthcoming August 2010 draft); Trial Tr. 2484:20-2487:14 (Demain) (discussing draft term sheets).

181. In December 2009, Pharmasset sent a draft term sheet to Merck which provided that Merck would grant Pharmasset a co-exclusive, worldwide license under Merck's patents with respect to the licensed compound, which was sofosbuvir. EX-1625 at 2; Trial Tr. 2486:9-20 (Demain).

182. In April 2010, Pharmasset sent a term sheet to Merck that provided for a similar license to Merck's patents. EX-1630; Trial Tr. 2486:25-2487:14 (Demain). Although these term sheets did not specifically mention the '499 and '712 Patents by name, the parties contemplated that Pharmasset would get a license to all of Merck's patents in this space. Trial Tr. 1412:16-1413:17 (Demain) (explaining that Pharmasset was looking to license "all of the patents related to HCV that Merck had"). At the time of these term sheet exchanges in late 2009 and 2010, the '499 Patent had issued and the application that led to the '712 Patent was pending with the Patent Office. EX-0001; EX-0002. And although the term sheets discussed were general in nature and did not list out the particular Merck patents that would have been licensed to Pharmasset, a final agreement would provide an appendix listing the licensed patents and patent applications. Trial Tr. 2507:18-24 (Demain).

183. Consistent with Pharmasset's repeated requests, a May 25, 2010, internal Merck

presentation about the Pharmasset term sheet indicated that Pharmasset had requested a "[n]on-exclusive, worldwide license under Merck patent rights and know how to develop, manufacture and commercialize products containing Licensed Compound [which included PSI-7977, Pharmasset's compound number for sofosbuvir]." EX-1634 at 3; Trial Tr. 2487:18-2489:3 (Demain).

184. On June 16, 2010, Merck sent Pharmasset a counter-proposal that did not include a license from Merck to Pharmasset that would provide Pharmasset freedom-to-operate with regard to Pharmasset's HCV products. EX-1636; Trial Tr. 1413:18-1414:7 (Demain) (explaining Pharmasset's proposed license was too broad and that Merck "took it out of the term sheet").

185. On August 5, 2010, Pharmasset wrote Merck in advance of sending a revised term sheet that once again sought a license to Merck's patent estate. The letter noted that "[t]he licensing of Merck Patent Rights and Know-How is specific to the development, manufacture and commercialization of PSI-7977 as a Monotherapy Product, or as the PSI-7977 component of Pharmasset Combination Products." EX-1652. While most of the term sheets exchanged during this period did not provide for a royalty to Merck, "there was one version that did have royalties going back to Merck." EX-1625 at 7; Trial Tr. 2506:23-2507:1 (Demain).

186. Around September 2010, Merck's interest in a deal changed from a collaboration to a purchase. On September 3, 2010, Merck again sent a letter that stated that one of the benefits to Pharmasset of an acquisition by Merck would include "'[t]he ability to leverage Merck's intellectual property estate to reduce uncertainty and enhance the value of the Pharmasset assets going forward.'" EX-0069; EX-0686 at 1-2; Trial Tr. 1414:14-1415:10 (Demain). Merck ultimately did not purchase Pharmasset.

187. In 2011, Merck executives informed Pharmasset's CEO, P. Schaefer Price, that Pharmasset needed a license from Merck to the '499 Patent to commercialize PSI-7977 (sofosbuvir). Merck indicated that "there were claims [of the '499 Patent] that could give Pharmasset trouble in the future." Mr. Price responded that he hoped Merck's attorney could "find the courthouse." Price Depo Tr. (EX-2392) at 115:13-116:06. This course of events is entirely inconsistent with a waiver of patent rights and demonstrates that Pharmasset did not hold any

belief—much less a reasonable one—that Merck had waived its patent rights.

188. The May 2011 Merck-Roche license, to which Pharmasset consented, is also inconsistent with a waiver. When Merck did not do a deal with Pharmasset for PSI-6130 in 2004, Pharmasset ultimately did a deal with Roche. EX-0627; Trial Tr. 1415:19-1416:4 (Demain). In 2011, when PSI-6130 was in phase II clinical studies and appeared as though it would advance to the next stage of development, Roche approached Merck for an unblocking license so that Merck's patents would not stand in the way of Roche bringing PSI-6130 (then renamed RG-7128) to the market. Trial Tr. 1416:9-23 (Demain). Pharmasset remained the development partner of that product with Roche. Trial Tr. 1417:14-20 (Demain). There is no evidence that Pharmasset ever conveyed to Roche that it thought that Merck was not going to enforce its patents against them.

189. In 2011, Roche (Pharmasset's development partner with regard to certain nucleosides including PSI-6130) entered into a license agreement with Merck, whereby Merck granted Roche a license to the '499 Patent (and other to-be-issued patents including the application that issued as the '712 Patent) and Roche agreed (among other things) to pay Merck a royalty of between 9-12%. EX-1783; Trial Tr. 1416:24-1417:7 (Demain).

190. Under Roche's development agreement with Pharmasset, Pharmasset's consent to the Roche-Merck license was sought because Roche's royalty payments to Merck would reduce Roche's royalty payments to Pharmasset. EX-0627 at 2; Trial Tr. 1417:18-1418:2 (Demain).

191. By September 7, 2011, Pharmasset had consented to the Roche-Merck license. EX-2632. Pharmasset was informed that Pharmasset's consent to the Merck-Roche license would cause the Merck-Roche license to spring into effect. EX-0619; Trial Tr. 1419:18-1423:1 (Demain). There is no evidence that Pharmasset ever told Roche that Merck would not assert its patents.

192. During the 2008 to 2011 timeframe, there is no evidence that anyone from Merck communicated to Pharmasset that Merck would not assert its patents. No one from Pharmasset ever communicated to Merck that Pharmasset thought Merck waived its patent rights. Trial Tr. 2482:2-18 (Demain).

193. In February 2, 2011, Merck prepared an internal business analysis that compared

two scenarios: one in which Merck would provide a license to Roche to develop product R-7128 and another in which Merck would buy Pharmasset and develop sofosbuvir. Trial Tr. 2514:11-2516:25 (Demain). The ’499 patents are listed as intellectual property considerations for the Roche license deal, but not for the Pharmasset sofosbuvir purchase deal. EX-0099 at 27, 29. But Ms. Demain explained this difference: in the first scenario (in which Roche would have to pay for a license to Merck’s patents), Merck was not contemplating a purchase of Roche; in the second scenario, in which Merck would buy Pharmasset, Merck’s patents would no longer be a concern for sofosbuvir—the only concern would be third-party patents. Trial Tr. 2516:3-25 (Demain) (explaining why Merck’s patents were listed on the R-7128 slide, but not the PSI-7977 slide). Ms. Demain’s testimony was not contradicted at trial and in any event, there is no indication that this document or any other like it was ever communicated to Pharmasset before this litigation commenced.

194. Merck had no viable patent infringement claim until Pharmasset/Gilead’s product was on the market. Trial Tr. 2483:3-7 (Demain). Given that Merck could not sue for infringement until late 2013 because Gilead’s pre-commercialization work is specifically exempted from constituting infringement under the “FDA exemption,” no ripe claim existed until then, and it would not be reasonable to conclude that Merck waived its patent rights before Gilead commercialized. Indeed, the ’712 Patent did not issue until the summer of 2013. EX-0002. Shortly thereafter, and before Gilead’s product was launched, Merck sent a letter to Gilead asking Gilead to take a license. EX-2566.

195. Furthermore, a defense of waiver cannot be asserted based on any interaction between Merck and Pharmasset in 2004 because Merck’s ’499 patent did not issue until September 12, 2006. EX-0001.

196. Gilead Response to Merck’s Interrogatory No. 11 (asking for the factual and legal basis for Gilead’s defense that Merck’s claims are barred by the equitable doctrine of laches and/or estoppel and/or waiver) does not point to any specific communications between Merck and Pharmasset, nor does Gilead’s response specify any document that indicates Merck has waived its right to assert the ’499 and ’712 Patents against Gilead. Gilead’s Written Discovery Responses 4-

5, ECF 231-25.

197. Gilead's Interrogatory response points only to EX-2314 as alleged evidence that Merck delayed assertion of its patent rights was misleading to Gilead or that Gilead has suffered material prejudice. Gilead's Written Discovery Responses 4-5, ECF 231-25. This reliance is misplaced: EX-2314 is a letter from Merck to Pharmasset dated September 3, 2010 regarding the *licensing proposal provided to Merck by Pharmasset*. The letter rejects the licensing proposal and rather suggests the alternative that Merck acquire Pharmasset.

198. Contrary to Gilead's assertion, EX-2314 specifically put Pharmasset on notice that Merck would assert its patent rights. In describing the benefits to Pharmasset and its shareholders in an acquisition of Pharmasset by Merck, the letter states that one of the benefits is "[t]he ability to leverage Merck's intellectual property estate *to reduce uncertainty and enhance the value of the Pharmasset assets* going forward." EX-2314 at 2 (emphasis added). The very document cited by Gilead shows that Merck communicated to Pharmasset that Merck's intellectual property estate was a source of uncertainty for Pharmasset.

199. No witnesses from either Pharmasset or Gilead testified that they reasonably believed that Merck would not assert its patents.

IV. CONCLUSIONS OF LAW - WAIVER

Courts have recognized waiver as a defense to patent infringement. *Qualcomm Inc. v. Broadcom Corp.*, 548 F.3d 1004, 1019 (Fed. Cir. 2008). There are two forms of waiver—"true waiver" and "implied waiver." *Id* at 1020. True waiver occurs when a patentee "with full knowledge of the material facts, intentionally relinquished its rights to enforce [the asserted patents]." *Id*. Implied waiver occurs when a patentee's "conduct was so inconsistent with an intent to enforce its rights as to induce a reasonable belief that such right has been relinquished." *Hynix Semiconductor Inc. v. Rambus Inc.*, 645 F.3d 1336, 1348 (Fed. Cir. 2011); *Qualcomm*, 548 F.3d at 1020.

In this case, Gilead does not contend that there was a true waiver of Merck's patent rights and instead argues Merck impliedly waived its patent rights. *See* Gilead Trial Br. 11-12, ECF 368. However, most courts finding an implied waiver of patents rights have done so in the context of

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standard setting organizations where (1) the patentee had a duty of disclosure to the standard setting organization and (2) the patentee breached that duty. *Barnes & Noble*, 849 F. Supp. 2d at 941-42 (citing *Hynix*, 645 F.3d at 1348); *see also Qualcomm Inc. v. Broadcom Corp.*, 2007 WL 1031373, at *6-23 (S.D. Cal. Mar. 21, 2007), *aff'd* 548 F.3d at 1020-22.

Gilead has cited three cases for the proposition that implied waiver is not limited to standard setting organizations. In *Mars, Inc. v. TruRX LLC*, the Eastern District of Texas discussed the Federal Circuit's decision in *Qualcomm*, which dealt with implied waiver in the standard setting context. Case No. 6:13-cv-526-RWS, ECF 346, at *2-3 (E.D. Tex. April 29, 2016). The Court found that "nothing in the [Federal Circuit's] opinion indicated that implied waiver can only be established if a patentee is under a duty to disclose information to a standard setting organization" and noted that "the [Federal Circuit] simply held that under the particular facts of the case, the district court did not abuse its discretion by concluding that Qualcomm's 'conduct was so inconsistent with an intent to enforce its rights as to induce a reasonable belief that such right ha[d] been relinquished.'" *Id*. at *2. What mattered to the court was not whether a standard setting organization was implicated, but rather whether the patent holder's silence or inaction was so inconsistent with an intent to enforce its rights as to induce a reasonable belief that the patent holder had relinquished its rights.

In *Universal Electronics Inc. v. Logitech, Inc.*, the Central District of California stated that "implied waiver as a doctrine does not need to be limited to" the context of a standard setting organization. Case. No. 11-cv-01056-JVS(ANx), ECF 144, at *21 (C.D. Cal. May 9, 2012). However, the court went on to recognize that it was aware of "no law dictating that silence outside of the [standard setting organization] context is 'so inconsistent' with intent to enforce" that it could constitute an implied waiver. *Id.* at *22. The court further recognized that "other courts" had "impos[ed] significant barriers to establish a duty to disclose in the [standard setting organization] context." *Id.*

In *Dane Technologies, Inc. v. Gatekeeper Systems, Inc.*, the final case relied upon by Gilead, the District of Minnesota appeared to assume that implied waiver is a valid defense outside the context of standard setting organizations. Case No. 12-cv-2730-ADM/JJK, 2015 WL 5719142, at *19 (D. Minn. Sept. 29, 2015). However, the court only cited cases involving standard setting organizations, and it did not analyze whether implied waiver could apply outside that context—it simply assumed so. *Id*.

While some courts have recognized implied waiver of patent rights outside the standard setting context, it is not clear that Federal Circuit caselaw dictates such a result. Assuming that implied waiver is a cognizable defense outside the standard setting context, Gilead has failed to meet its burden of proof. On that note, it is also unclear whether the burden of proof for asserting waiver is preponderance of the evidence or clear and convincing evidence. *See, e.g. Hynix*, 645 F.3d 1348 ("To support a finding of implied waiver in the standard setting organization context, the accused must show by clear and convincing evidence…") (quoting *Qualcomm*, 548 F.3d at 1020); *A.C. Aukerman Co. v. R.L. Chaides Construction Co.*, 960 F.2d 1020, 1045-46 (Fed. Cir. 1992) (en banc) (holding that the quantum of proof for equitable estoppel is a preponderance of the evidence except where "special considerations" are implicated, such as "where the danger of deception is present . . . , where a particular claim is disfavored on policy grounds . . . , or where a particularly important individual interest is at stake such as one's reputation . . . ."); *Oracle Am., Inc. v. Google Inc.*, Case No. 10-cv-03561 WHA, 2012 WL 1965778, at *2 (N.D. Cal. May 31, 2012) ("To prevail on a waiver defense, Google must show by a preponderance of the evidence…"). For purposes of this case, the Court need not decide the issue as Gilead has failed to prove implied waiver by either standard of proof.

Implied waiver requires proof that the patentee's conduct "was so inconsistent with an intent to enforce its rights as to induce a reasonable belief that such right has been relinquished." *Hynix*, 645 F.3d at 1348 (quoting *Qualcomm*, 548 F.3d at 1020)); *see also* Pretrial Conference Statement 5, ECF 254 (stipulation that waiver requires "a reasonable belief that [a] right has been relinquished"). Gilead has failed to make such a showing for at least three reasons:

First, Gilead failed to establish that it or Pharmasset reasonably believed that Merck had relinquished its patent rights. Gilead did not offer any evidence to show such a belief. In fact, the only evidence of what Pharmasset or Gilead believed supports a conclusion that they did not believe Merck had relinquished its rights. *See supra*, FOF ¶¶ 179, 187. This failure of proof alone compels a conclusion that implied waiver has not been shown.

Second, even if Gilead had offered evidence tending to show that Pharmasset or Gilead believed Merck had relinquished its right to assert the patents in suit, any such belief would have been unreasonable because Merck's conduct was not inconsistent with an intent to enforce its rights. From 2008 to 2011, the parties engaged in repeated discussions over partnership opportunities in the antiviral space. During such discussions, Pharmasset proposed term sheets to Merck which provided that Merck would grant Pharmasset a worldwide license to Merck's patents. In one counter-proposal, Merck sent an offer that did not provide Pharmasset with a freedom-to-operate license with respect to Pharmasset's HCV products. Furthermore, at a meeting in 2011 in which Merck informed Pharmasset that the '499 patent "could give Pharmasset trouble in the future," Mr. Price told a Merck attorney that he "hoped [the Merck attorney] found it easier to find the courthouse." *See supra*, ¶ 189. Such conduct would not create a reasonable belief that Merck had relinquished its rights to enforce the asserted claims. Gilead's attempt to characterize these negotiations as fundamentally inconsistent with an intent to enforce patent rights glosses over several facets of the negotiations. For example, Gilead claims in 2010 that Merck never told Pharmasset that Pharmasset should offer it different terms because Merck had patents that covered PSI-7977. However, in 2010, Merck responded to Pharmasset's proposals with counter-offers that did not provide a license for Pharmasset's HCV products. This is not the conduct of a party (Merck) that had waived its right to enforce its patents or of a party (Pharmasset) that has a "reasonable belief" that Merck had waived its patent rights.

Finally, it does not appear that Merck had an actionable claim of infringement until Gilead's product was launched on the market in December 2013. Gilead's development activities prior to the launch is protected from infringement liability under 35 U.S.C. § 271(e)(1). *See generally Merck KGaA v. Integra Lifesciences I, Ltd.*, 545 U.S. 193, 202 (2005) (explaining the § 271(e)(1) safe harbor). Since Merck could not enforce its patents until Gilead's product launched, Merck had no affirmative duty to take any action and its failure to take any action cannot be interpreted as implied waiver. *See, e.g., Bio-Tech. Gen. Corp. v. Genentech, Inc.*, 80 F.3d 1553, 1564 (Fed. Cir. 1996) (holding in the context of laches that "[w]ith no legal right to enforce, it cannot be said that Genentech unreasonably delayed during that time period [before FDA approval and launch].").

The Court concludes that Gilead has not proven its waiver defense and that Merck is not prohibited from asserting its patents on this basis.

V. CONCLUSIONS OF LAW – UNCLEAN HANDS

A. Background on Unclean Hands

The equitable doctrine of unclean hands has long existed as a principal of patent law. It arises from the maxim, "[h]e who comes into equity must come with clean hands." *Keystone Driller Co. v. Gen. Excavator Co.*, 290 U.S. 240, 241 (1933). The party asserting the defense of unclean hands must prove it by clear and convincing evidence. *In re Omeprazole Patent Litig.*, 483 F.3d 1364, 1374 (Fed. Cir. 2007); *Aptix Corp. v. Quickturn Design Sys., Inc.*, 269 F.3d 1369, 1374 (Fed. Cir. 2001). In a trio of cases in the 1930s and 1940s, the Supreme Court applied the doctrine of unclean hands to dismiss patent cases involving egregious misconduct.

First, in *Keystone*, which involved the manufacture and suppression of evidence, the plaintiff sued for patent infringement. 290 U.S. at 242. In an earlier infringement action against a different defendant, Keystone had prevailed and its three patents were declared valid. *Id.* Armed with this verdict, Keystone brought suit against the General Excavator Company and another company for infringing the same three patents and moved for a preliminary injunction. *Id.* The injunction was denied, and Keystone amended its complaint to allege infringement of two more patents. *Id.* The case then proceeded to trial. *Id.* at 242-43.

During the trial, it was discovered that after learning about a possible invalidating prior use, the patent applicant, who was Keystone's general manager and secretary, for one of the patents-in-suit paid the potential prior user to sign a false affidavit stating the prior use was an abandoned experiment, to assign any rights to the applicant, and to suppress any evidence of the prior use. *Id.* at 243. The Supreme Court framed this issue on appeal as follows:

> Plaintiff contends that the [unclean hands] maxim does not apply unless the wrongful conduct is directly connected with and material to the matter in litigation, and that, where more than one cause is joined in a bill and plaintiff is shown to have come with unclean hands in respect of only one

> of them, the others will not be dismissed.

Id. at 244. The Supreme Court described the general doctrine of unclean hands:

> [Plaintiff] must come into court with clean hands. He must be frank and fair with the court, nothing about the case under consideration should be guarded, but everything that tends to a full and fair determination of the matters in controversy should be placed before the court…It is a principle in chancery, that he who asks relief must have acted in good faith. The equitable powers of this court can never be exerted in behalf of [one] who has acted fraudulently, or who by deceit or any unfair means has gained an advantage. To aid a party in such a case would make this court the abetter of iniquity.

Id. at 244-45 (internal quotations and citations omitted). With that in mind, the Supreme Court explained that unclean hands applies only where the "unconscionable act of one coming for relief has immediate and necessary relation to the equity that he seeks in respect of the matter in litigation." *Id.* at 245. The misconduct must "affect the equitable relations between the parties in respect of something brought before the court for adjudication." *Id.* In *Keystone*, the Supreme Court stated that "it [] clearly appear[ed] that [Keystone] made the [first] case a part of his preparation in the [subsequent suits]." Therefore, Keystone's conduct with respect to one patent was sufficient to infect causes of action based on related patents and to prevent recovery on any of the asserted patents. *Id.* at 247.

Second, in *Hazel-Atlas Glass Co. v. Hartford-Empire Co.*, 322 U.S. 238 (1944), overruled on other grounds by *Standard Oil Co. v. United States*, 429 U.S. 17 (1976), also involving the manufacture and suppression of evidence, Hartford alleged Hazel-Atlas infringed its patent. The District Court, finding that infringement had not been proven, dismissed the case. *Id.* at 241. On appeal, the Circuit Court, quoting extensively from an article written by William Clarke, an expert and former President of the Glass Workers' Union, found the patent valid and infringed. *Id.* at 241-42. The Circuit Court's decision caused both Hazel-Atlas and Hartford to contact Mr. Clarke, who eventually signed an affidavit that he wrote the article. *Id.* at 242-43. Hazel-Atlas then settled the patent lawsuit with Hartford. *Id.* at 243. In a separate anti-trust action by the United States against Hartford, seven years after the patent dispute, evidence disclosed that the patentee's attorney wrote the article to overcome issues at the Patent Office and had Mr. Clarke sign it as his own and publish it. *Id*. at 243-44.

The Supreme Court explained that the doctrine of unclean hands "has always been characterized by flexibility which enables it to meet new situations which demand equitable intervention, and to accord all the relief necessary to correct the particular injustices involved in these situations." *Id*. at 248. In *Hazel-Atlas*, the Court found the fraud was so egregious that it found the patent unenforceable against Hazel-Atlas and denied any recovery. *Id*. at 249-251.

Third, in *Precision Instrument Manufacturing Co. v. Automotive Maintenance Machinery Co.*, 324 U.S. 806 (1945), involving perjury and suppression of evidence, Automotive sued Precision for breach of contract and patent infringement. The parties had been adversaries in a prior interference proceeding, with competing patent applications covering torque wrenches. *Id*. at 809-12. During the interference proceeding, Automotive learned that Precision filed a fraudulent affidavit. *Id*. Instead of reporting this fraud to the Patent Office, Automotive settled the interference case with Precision and Precision assigned its rights in the application to Automotive. *Id*. When Precision recommenced selling the allegedly infringing torque wrenches, Automotive brought suit against Precision. *Id*. at 814.

The Supreme Court reiterated general principals of the doctrine of unclean hands, including the broad discretion an equity court has in refusing to be an accomplice to the unclean litigant. *Id*. at 815. Commenting that "the maxim is far more than a banality," the Court explained:

> [The maxim of unclean hands] gives wide range to the equity court's use of discretion in refusing to aid the unclean litigant. It is "not bound by formula or restrained by any limitation that tends to trammel the free and just exercise of discretion." Accordingly one's misconduct need not necessarily have been of such a nature as to be punishable as a crime or as to justify legal proceedings of any character. Any willful act concerning the cause of action which rightfully can be said to transgress equitable standards of conduct is sufficient cause for the invocation of the maxim by the chancellor. Moreover, where a suit in equity concerns the public interest as well as the private interests of the litigants this doctrine assumes even wider and more significant proportions. The possession and assertion of patent rights are "issues of great moment to the public."

Id. at 815 (internal citations omitted).

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The Supreme Court found that the history of the patents-in-suit was steeped in perjury and undisclosed knowledge of perjury. *Id.* at 816. The Court neither found nor required a finding that any of the patents-in-suit would not have issued if Automotive had disclosed to the examiner the information provided by its former employee. *Id.* at 815-19. Moreover, that information plainly had no bearing whatever on the patents that issued from Automotive's own applications. *Id.* Yet the Court ruled that Automotive's unclean hands prevented enforcement of all of the patents-in-suit. *Id.* at 819.

Notably, in *Hazel-Atlas* and *Precision*, the Supreme Court reversed lower courts that had been unwilling to bar suit for the described misconduct. In *Keystone*, the circuit court reversed the district court's finding denying the unclean hands defense which was affirmed by the Supreme Court.

Almost 70 years after *Precision*, the Federal Circuit issued its *en banc* decision in *Therasense, Inc. v. Becton, Dickson & Co.*, 649 F.3d 1276 (Fed. Cir. 2011). *Therasense* addressed the separate defense of inequitable conduct—a defense that Gilead does not assert in this case—but the Federal Circuit's discussion of the differences between inequitable conduct and unclean hands confirmed that unclean hands remains a viable defense to patent infringement. *Id.* at 1285-89. As the Federal Circuit explained, the doctrine of inequitable conduct grew from the older doctrine of unclean hands. *Id.* at 1287. Whereas unclean hands can involve improper conduct before either the Patent Office or the courts, inequitable conduct relates solely to conduct before the Patent Office. *Id.* Additionally, where unclean hands affects the enforceability of a patent in a particular lawsuit, inequitable conduct carries far more severe consequences for the patent holder—"unenforceability of the entire patent rather than mere dismissal of the instant suit." *Id.* For this reason, inequitable conduct requires a "finding of both intent to deceive and materiality." *Id.* The Federal Circuit made clear, however, that unclean hands remains a viable defense, and does not require a finding of materiality:

> This court recognizes that the early unclean hands cases do not present any standard for materiality. Needless to say, this court's development of a materiality requirement for inequitable conduct does not (and cannot) supplant Supreme Court precedent. Though inequitable conduct developed

> from these cases, the unclean hands doctrine remains available to supply a remedy for egregious misconduct like that in the Supreme Court cases.

Id. Thus, the Federal Circuit's *Therasense* decision confirmed the continuing viability of the unclean hands doctrine.

B. Other Cases Involving Unclean Hands

Against this standard from the Supreme Court and Federal Circuit, other courts have applied the doctrine of unclean hands to situations involving lying under oath, unethical business conduct, or litigation misconduct.

In *Aris-Isotoner Gloves, Inc. v. Berkshire Fashions, Inc.*, 792 F. Supp. 969, 970 (S.D.N.Y. 1992), *aff'd*, 983 F.2d 1048 (2d Cir. 1992), the Court found egregious misconduct where the Defendant's president lied under oath in a prior proceeding. In an attempt to prove detrimental reliance on Plaintiff's conduct, Berkshire President Issac Dweck testified at a contempt hearing that his company initially sold very small quantities of an infringing glove and after nothing happened—it was not sued for infringement—the company increased the amounts sold in the following years. *Id.* In a remand hearing, after being confronted with contrary evidence in interrogatory responses, Mr. Dweck testified that Berkshire sold over 50,000 dozen gloves and sales decreased, not increased, the following year. *Id.* He also admitted that his prior testimony had been incorrect even though the relevant figures had been available to him at the prior hearing. *Id.*

The court found that Mr. Dweck had fabricated his testimony in light of "the inadequately explained and obvious contradictions as to testimony of direct relevance." *Id.* The court also rejected Berkshire's explanation that Mr. Dweck had confused sales of the infringing glove with another glove as "wholly inconsistent" with Mr. Dweck's "original, confident story." *Id.* at n.2. The court also rejected Berkshire's contention that Mr. Dweck's inconsistent testimony was immaterial because regardless of which version was believed, it did not affect the outcome. *Id.* at 971. However, the court found that once Berkshire engaged in the egregious misconduct, the doctrine of unclean hands prevented Berkshire from obtaining relief. *Id.* Other courts have also found unclean hands in the presence of false testimony. *See Mas v. Coca-Cola Co.*, 163 F.2d 505, 511 (4th Cir. 1947) (finding the plaintiff had unclean hands and upholding dismissal of plaintiff's suit where plaintiff submitted false testimony and forged documents to the Patent Office); *C.C.S. Commc'n Control, Inc. v. Sklar*, Case No. 86-cv-7191-WCC, 1987 WL 12085, at *2-3 (S.D.N.Y. 1987) (denying request for equitable remedy because plaintiff committed perjury).

Improper business conduct can also invoke unclean hands. In *Clements Indus., Inc. v. A. Meyers & Sons Corp.*, 712 F. Supp. 317, 318 (S.D.N.Y. 1989), plaintiff attempted to extract confidential information from the defendant, not for legitimate commercial reasons, but rather to obtain the defendant's confidential trade secrets. The court found that "[t]his deceptive dealing fully supports [defendant's] contention that [plaintiff] has 'unclean hands'" and dismissed plaintiff's claims. *Id*. at 328.

Courts have found improper business dealings can invoke unclean hands in several other situations. *See Worthington v. Anderson*, 386 F.3d 1314, 1321-22 (10th Cir. 2004) (affirming dismissal of plaintiff's trademark claims against former business partner for unclean hands where plaintiff "threw economic obstacles in the way of" defendant's ability to comply with terms of arbitration agreement); *Saudi Basic Indus. Corp. v. ExxonMobil Corp.*, 401 F. Supp. 2d 383, 395 (D.N.J. 2005) ("There is also caselaw to support application of the unclean hands doctrine when a business partner engages in acts of self-dealing."); *FLIR Sys., Inc. v. Sierra Media, Inc.*, 965 F. Supp. 2d 1184, 1197 (D. Or. 2013) ("FLIR's false advertising claim . . . is barred, in light of FLIR's false advertising on the same subject matter, by the doctrine of unclean hands."); *Unilogic, Inc. v. Burroughs Corp.*, 10 Cal. App. 4th 612, 617-621 (1992) (affirming, *inter alia*, that plaintiff's failure to return defendant's software and continued use of software after development agreement terminated was unclean hands barring plaintiff's legal claim for conversion); *Fed. Folding Wall Corp. v. Nat'l Folding Wall Corp.*, 340 F. Supp. 141, 146 (S.D.N.Y. 1971) (plaintiff breaching employment contract with defendant and inducing trademark owner to cancel license to defendant was unclean hands warranting dismissal of case); *Metro Publishing, Ltd. v. San Jose Mercury News, Inc.*, 861 F. Supp. 870, 880 (N.D. Cal. 1994) (finding plaintiff's deliberate attempt to create trademark confusion constituted unclean hands and granting summary judgment against trademark holder "on this basis alone").

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Courts have also found unclean hands applicable where a party has engaged in litigation misconduct. In *U.S. Ethernet Innovations, LLC v. Texas Instruments Inc.*, Case No. 6:11-cv-491-MHS, 2014 WL 4683252, at *6 (E.D. Tex. 2014), defendant's unprofessional conduct, including attempting to interfere with plaintiff's expert, constituted unclean hands

C. Application of Unclean Hands to Findings of Fact

Against this backdrop, the Court must review the facts to determine whether Merck's misconduct rises to the level of egregious misconduct sufficient to bar Merck from maintaining this suit against Gilead. All of the Court's findings are made under the standard of clear and convincing evidence.

In this case, numerous unconscionable acts lead the Court to conclude that the doctrine of unclean hands bars Merck's recovery against Gilead for infringement of the '499 and '712 Patents. Merck's misconduct includes lying to Pharmasset, misusing Pharmasset's confidential information, breaching confidentiality and firewall agreements, and lying under oath at deposition and trial. Any one of these acts—lying, unethical business conduct, or litigation misconduct—would be sufficient to invoke the doctrine of unclean hands; but together, these acts unmistakably constitute egregious misconduct that equals or exceeds the misconduct previously found by other courts to constitute unclean hands. Merck's acts are even more egregious because the main perpetuator of its misconduct was its attorney.

1. Pharmasset and Merck Interactions

The first set of unconscionable acts barring Merck's recovery from Gilead for infringement concerns the actions of Merck and its patent prosecutor, Dr. Durette, in learning the confidential structure of Pharmasset compound PSI-6130 and pursuing patent claims to cover that compound in violation of the Merck-Pharmasset firewall and Merck's own policies.

Interactions between Merck and Pharmasset began in 2001 when the companies discussed potential collaboration opportunities. FOF ¶ 37. As part of these discussions, the companies signed a NDA. *Id.* In 2003, pursuant to the NDA, Pharmasset gave Merck an overview of its HCV program, including an overview of its lead compound, PSI-6130. FOF ¶¶ 42-44. Shortly after, the companies signed a Material Transfer Agreement (MTA), which permitted Merck to test and evaluate PSI-6130. FOF ¶ 46. After the testing revealed encouraging results, Merck requested additional information about the structure of PSI-6130. FOF ¶ 50. Merck assured Pharmasset that structural information about PSI-6130 would be firewalled and on this basis, the parties set up a phone call for March 17, 2004. FOF ¶¶ 53-59.

It was not as though Merck and Dr. Durette stumbled into that call unaware of the subject matter, or the impropriety of Dr. Durette's participation. All of this information was contained in emails and a term sheet distributed to Merck, and Dr. Durette in particular, in advance of the meeting. In these e-mails, Merck's employees were fully advised in advance that Pharmasset would disclose its closely guarded PSI-6130 compound to Merck employees bound by an NDA and firewall. Merck further knew that Pharmasset's compound was an NS5B polymerase inhibitor just like its own compounds from the Merck-Isis collaboration that formed the bases of the '499 and '712 patent applications. Dr. Durette's legal and scientific sophistication preclude the possibility that he was unaware or misunderstood the relationship of the anticipated disclosure to his own HCV work for Merck.

Compounding the problem, Merck's representatives, Dr. Durette and Dr. Pon, committed further unconscionable acts during the call. Based on the contemporaneous notes prepared by Pharmasset's Alan Roemer, after learning key structural features of PSI-6130, Dr. Durette voiced concern that he might have a problem, stating "seems quite related to things I'm involved with," EX-2098, but he never revealed that he was prosecuting Merck's own HCV patent applications. This was information unavailable to Pharmasset. Moreover, Dr. Durette's involvement with Merck's HCV patents violated the understanding the parties had about their firewall obligations, which excluded anyone involved with Merck's internal HCV program. EX-2302. This most certainly would include the Merck-Isis collaboration that Dr. Durette was involved with. After suggesting there might be a problem, both Dr. Durette and Dr. Pon assured Pharmasset that they were within the firewall and continued the conversation.

On that call, Dr. Durette obtained the full structure of PSI-6130 and he subsequently continued to prosecute Merck's HCV patent portfolio. Although he claims to have recused himself from the Pharmasset-Merck due diligence, that is not where the harm lay. It was, in fact, wrong for Merck to allow Dr. Durette to continue to prosecute the ’499 and ’712 Patent applications. Ironically, in the course of what the Court deems a complete fabrication of testimony at his deposition, Dr. Durette himself explained why this conduct was so egregious. As he said, having learned the structure of PSI-6130, his judgment was tainted. And, indeed it was. His February 2005 claim amendments to the ’499 patent were made possible by the information he unfairly obtained in March 2004. Proper recusal would have mandated that Dr. Durette cease work on Merck’s HCV patents as well. Such conduct was required by Merck’s own internal policies and would have been consistent with a common understanding of recusal.

Based on the foregoing, there can be no doubt that Merck used this highly confidential information to benefit its own prosecution of its stalled ’499 Patent application. Dr. Pon and Dr. Durette’s deception about Dr. Durette being firewalled, and Merck’s subsequent decision to allow Dr. Durette to continue to prosecute the ’499 and ’712 with full knowledge of the structure of Pharmasset’s PSI-6130 constitute unacceptable business conduct. It is clear to this Court that Dr. Durette improperly used this information to inform his conduct in amending the ’499 Patent claims a mere 18 days after the Clark application published. Those amendments related to compounds Merck never tested during its collaboration with Isis, and the amendments were not prompted by requests from the inventors or prodding by the patent examiner to narrow the claim scope. Thinking that he was now free from what he knew were his obligations under the NDA, Dr. Durette pounced on the opportunity to capitalize on what he improperly had learned a year earlier.

The Court concludes that each of the foregoing unconscionable acts has an “immediate and necessary relation to…the matter in litigation” because the patents that resulted from this series of unconscionable acts are now asserted against Gilead, Pharmasset’s successor-in-interest. *See Keystone*, 290 U.S. at 245. The Court finds the facts in *Clements* analogous to Merck’s misconduct. In *Clements*, the court found plaintiff’s deceptive dealing in learning defendant’s confidential trade secrets warranted a finding of unclean hands. In a similar situation, Merck sent Dr. Durette to “view the structure during a patent due diligence meeting” under deceptive circumstances. EX-0153.0001. As detailed *supra* FOF ¶¶ 54-92, the evidence shows Dr. Durette

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lied to Pharmasset about being within the firewall, then Merck allowed Dr. Durette, with his tainted judgment, to continue prosecuting the related Merck-Isis patents-in-suit and to draft claims to target Pharmasset's inventions. The Court finds Merck's deceptive dealing warrants a finding of unclean hands. *See Clements*, 712 F. Supp. at 328.

2. Litigation Misconduct

The Court concludes that the doctrine of unclean hands also bars Merck's recovery against Gilead for infringement of the '499 and '712 Patents based on additional reprehensible acts by Merck and Dr. Durette amounting to litigation misconduct, including his false testimony in this case. Based on the Court's findings *supra* FOF ¶¶ 107-170, the record shows that Dr. Durette presented inconsistent, contradictory, and untruthful testimony, and that testimony was sponsored by Merck.

Throughout the prosecution of this case, Dr. Durette continued to deceive Gilead and this Court. His trial testimony was inconsistent with his deposition testimony in numerous material and critical respects. He recanted a major portion of his prior testimony without any warning to Gilead until revealed in Merck's opening statement.[3] He gave inconsistent stories about his participation on the March 2004 due diligence call and the circumstances that led to his amendments to the '499 claims. His trial testimony was not credible on significant matters related to this case.

Remarkably, when he faced the Court and jury at trial, Dr. Durette recanted his testimony that he had not been on the Pharmasset-Merck due diligence call. At trial, he testified that he just did not remember what had taken place 11 years ago. Trial Tr. 347:9-22 (Durette). His trial testimony is completely inconsistent with his deposition testimony. Dr. Durette had previously testified at his deposition that he was certain he had not participated in the call and not learned the structure of Pharmasset's compound:

> Q: How can you be so sure 11 years later that you were never told what

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[3] Also troubling is Merck's counsel's failure to disclose to Gilead or this Court that Dr. Durette would recant his prior testimony as soon as Merck learned that Dr. Durette's prior testimony was unsustainable—wholly inconsistent with the record evidence. Opening statement was not the preferred time for such a disclosure. *See* ABA Model Rules Prof. Conduct, Rule 3.3(a).

> the structure was for the 6130 compound?
>
> A: The structure was not revealed to me by individuals at Merck or otherwise. I'm positive of that. I never saw a structure of the Pharmasset compounds until it was published later on in time."

Durette Dep. Tr. (EX-2388) at 31:4-10.

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> Q: How do you know you weren't told it?
>
> A: Because I remember that.
>
> Q: You remember what?
>
> A: That the structure was not disclosed to me
>
> Q: How do you remember that?
>
> A: Because I do.

Durette Dep. Tr. at 169:10-18, ECF 410-3.

Further, as rationale for his memory of the events, Dr. Durette embellished his "clear" recollection during his deposition by stating confidently—even sanctimoniously:

> Q: How can you be so sure of that memory?
>
> A: Because I was not part of the patent due diligence for the structure, so I would not have been privy to any revelation of the structure to me as a patent attorney working on a related docket. So this was assigned to another person. I would not have participated in a phone call wherein it was a potential for the revelation of the structure to Merck counsel.
>
> Q: Why would that have been inappropriate for you to have been told the structure of 6130?
>
> A: Because I was prosecuting a docket which had potential a conflict with Pharmasset's IP positions on the subject matter.

Durette Dep. Tr. (EX-2388) at 38:1-13.

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> Q: Again, why would it have been inappropriate or wrong for you to have

> been told the 6130 structure?
>
> A: It would have tainted my judgment as to what claims to pursue in the Merck/Isis collaboration.
>
> Q: How would it have tainted your judgment?
>
> A: Having structural information is very important as to what the competition is doing in its research efforts. We had a policy in Merck on a particular docket area if there were potential licensing opportunities in a related area, that due diligence would be assigned to a non—an attorney that was not prosecuting a particular docket in a related area.

Durette Dep. Tr. (EX-2388) at 38:21-39:7.

Dr. Durette's trial testimony about failed memory rings hollow. By the time he appeared at trial, Dr. Durette was aware that Pharmasset's Alan Roemer had contemporaneous notes that indisputably placed him at the meeting and would expose his false testimony. But that was not the end of Merck's problems. As he tried to put a new gloss on his conduct, Dr. Durette placed blame on his colleague Pamela Demain, stating that she had instructed him to attend the due diligence call and that his supervisor approved it. However, Ms. Demain testified credibly that she did not.

He further testified untruthfully that before the meeting he had "no knowledge of what the structure was going to be revealed to me." Trial Tr. 351:3-4 (Durette). He stated that he and his supervisor concluded that there was little chance of overlap with Dr. Durette's HCV docket since the field of nucleosides was so broad. However, this testimony simply does not hold up against the information about Pharmasset's compound disclosed on the term sheet that Merck and Dr. Durette reviewed before the meeting. As Ms. Demain credibly testified, Merck knew going into the meeting that Pharmasset's compound was an NS5B polymerase inhibitor just like Merck's compounds. Moreover, it is not credible to the Court that Dr. Durette had such a clear memory about a meeting with his supervisor prior to the due diligence call when he also testified that he lacked any memory of the events 11 years prior.

Further at trial, Dr. Durette spun a new tale about the genesis of the February 1, 2005, amendments to the claims in the '499 patent application. At his deposition, Dr. Durette could not

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recall when he had first seen the Clark patent application containing PSI-6130 that was published on January 13, 2004. He averred that he might not have seen it until after he filed his amended claims. Durette Dep. Tr. 51:2-15, ECF 410-3. He further testified that he did not associate the Clark patent application with PSI-6130; he explained:

> Q: How is it that you would know that you would not in January 2005 have realized that Paragraph 0168, that chemical structure there, was 6130?
>
> A: Because this was one compound out of a plethora of compounds in the publication.

Durette Dep. Tr. at 52:19-25, 53:1-6, ECF 419-1.

Although Dr. Durette professed not to recall seeing the Clark publication before his amended claims were filed, he did have a clear recollection of other publications that "pointed towards fluoro as being an important invention for HCV nucleosides…." Durette Dep. Tr. at 65:18-25, ECF 410-3. When asked at his deposition why he had amended the claims on February 1, 2005, he testified "We wanted to expedite prosecution of the application." Durette Dep. Tr. at 62:5-9, ECF 419-1. He also testified that competitors were disclosing fluoro compounds that Merck had support for in its patent applications. Durette Dep. Tr. at 63:18-64:7, ECF 419-1. However, he avoided associating his amendment with the Clark publication.

At trial, Dr. Durette offered different reasons for the amendments. He testified that in addition to wanting to expedite the examination, Merck wanted to capture the subject matter that was most important to the Merck-Isis collaboration. Trial Tr. 404:14-19 (Durette). This testimony was in stark contrast to the testimony of other witnesses that Merck had never tested any of those compounds during the Merck-Isis collaboration and none of the inventors had discussed the amendments with him before the amendment. Dr. Durette's testimony is not credible on this issue.

Additionally, at trial, Dr. Durette now recalled clearly that he did see the Clark publication before he filed the amendments. When asked when he recalled seeing the Clark publication, Dr. Durette testified:

> A: I don't have a specific recollection of the timing, but I know it was

> before the filing of my second amendment because of two reasons: A, I was monitoring the competition in the area, and B, there must have been a triggering event that led me to reexamine my docket and take a look at my ’499 application which had been pending for about a year and a half. So I was convinced – or I became convinced that it was the publication of the application that led me to reexamine and then file the secondary amendment, or secondary amendment 18 days later.

Trial Tr. at 390:25-391:9 (Durette).

Although Merck would ask this Court to accept the simple explanation that Dr. Durette’s memory failed him and that the inconsistencies are harmless, in light of Dr. Durette’s persistent pattern of falsifications, the Court cannot interpret his testimony in this manner. It is overwhelmingly clear to the Court that Dr. Durette sought at every turn to create the false impression that Merck’s conduct was above board.

Knowing that he should not have been on the Pharmasset call and that upon learning the structure of PSI-6130, Dr. Durette should have recused himself from the Merck HCV docket. Instead, he first tried to deny knowledge of his role in the Pharmasset due diligence. When that did not work, he recanted his sworn testimony at trial and tried to blame others at Merck for compelling him to participate in the call. In order to first justify the propriety of the claim amendments made on the heels of the Clark publication, first he claimed not to have seen the Clark publication before he filed his amendments and when that story did not pan out he testified at trial that the Clark publication was actually the trigger that caused him to reexamine his stale ’499 claims.

In sum, several important facts are clear. First, Dr. Durette provided false testimony to this Court on important issues regarding Merck’s validity claims. Second, Merck sponsored and encouraged Dr. Durette’s conduct in the prosecution of the ’499 Patent, including Dr. Durette’s improper participation on the Pharmasset call and his continued prosecution of Merck’s HCV docket. Third, Merck fully aligned itself with Dr. Durette, as evidenced by its provision of legal counsel to Dr. Durette at his deposition and trial and designation of him as a Rule 30(b)(6) witness on selected issues. Merck's counsel spent two days preparing him for his deposition and for trial. Fourth, the untruthful testimony offered by Dr. Durette in his deposition and at trial was not incidental, but rather was directed at and supported Merck's validity arguments, and went to the heart of significant issues in this case. Fifth, by making Dr. Durette a centerpiece of its case, from the opening statement to the closing argument, Merck's litigation misconduct infects the entire lawsuit, including the enforceability of the '712 Patent.

The Court concludes that Dr. Durette's testimony has an "immediate and necessary relation to . . . the matter in litigation" because Dr. Durette testified regarding the key invalidity defenses presented to the jury, and regarding how Merck obtained the patents that are now asserted against Gilead, Pharmasset's successor-in-interest. *Keystone*, 290 U.S. at 245. Dr. Durette's testimony played an influential role at trial on the critical issue of the relationship between the amended '499 claims drafted solely by Dr. Durette and the content of the earlier specification. In response to questions by Merck, he testified that the claims were fully described in the application he filed in 2002. *See supra*, FOF ¶ 135. Although other witnesses presented testimony regarding written description and enablement, Dr. Durette was a key witness on this issue and thus, such additional evidence does not absolve Merck of its unclean hands with respect to Dr. Durette's fabrications.

The Court finds the *Aris-Isotoner* case particularly persuasive as it relates to Merck's misconduct at Dr. Durette's deposition and at trial. In *Aris-Isotoner*, the defendant's president gave testimony in one proceeding that directly contradicted his testimony in a prior proceeding. 792 F. Supp. at 970. That court found "no other conclusion can exist but that [defendant's president] fabricated his testimony in either the instant proceedings or in the original contempt proceedings." *Id*. That court found the witness's "half-hearted" claim that he was "confused" in the initial proceeding was "wholly inconsistent with [his] original, confident story." *Id*. at 970 n.2. On the basis of the fabricated testimony, the court dismissed defendant's laches defense. Id. at 972. This Court finds these facts akin to Dr. Durette's confident explanation at his deposition, recanted at trial, about why he never learned the structure of PSI-6130 from Pharmasset and his wholly inconsistent testimony regarding the genesis of the February 1, 2005, claims amendments

As in *Aris-Isotoner*, Dr. Durette's deposition testimony and trial testimony in this case are irreconcilable. The Court concludes that Dr. Durette lied in both proceedings. Further borrowing from *Aris-Isotoner*, this Court "lack[s] complete confidence as to which—if either—of the two testimonies is correct." *Aris-Isotoner*, 792 F. Supp. at 971. The Court concludes that Dr. Durette's fabricated deposition testimony and his false trial testimony, both of which Merck sponsored, are unconscionable acts that warrant a finding of unclean hands.

The Court also takes into account the fact that Dr. Durette was Merck's attorney. Among many important duties, attorneys have a duty of candor.[4] The legal system requires witnesses to supply complete and truthful testimony. If a witness fabricates testimony, justice is not served and when an attorney lies under oath, the Court cannot sanction such conduct. Dr. Durette, as Merck's former employee and 30(b)(6) witness, lied repeatedly at his deposition and at trial. The Court cannot condone such conduct from any witness, let alone an attorney.

3. Merck's Arguments Against Unclean Hands

Merck argues that Gilead's theory of unclean hands is precluded by the jury's verdict. If it is not, Merck denies all misconduct and seeks to diminish Dr. Durette's testimony to the failed memory of a retired employee. Alternatively, Merck argues that even if the Court finds fabricated testimony, unethical business practices, and litigation misconduct, none of that conduct amounts to unclean hands for several reasons: (1) its misconduct is not egregious; (2) amending claims to cover a competitor's product is expressly allowed; (3) Merck and Dr. Durette did not have an intent to deceive; (4) Dr. Durette's conduct cannot be imputed to Merck; (5) there is no immediate and necessary relation between the alleged misconduct and the litigation; and (7) any misconduct did not involve the '712 Patent. The Court addresses each in turn.

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[4] The New Jersey Disciplinary Rules of Professional Conduct, Rule 3.3 which governs candor toward the tribunal, provides: "A lawyer shall not knowingly: (1) make a false statement of material fact or law to a tribunal." N.J. R.P.C. § 3.3(a)(1). Rule 4.1 governs truthfulness in statements to others, and provides: "In the course of representing a client a lawyer shall not knowingly: (1) make a false statement of material fact or law to a third person." N.J. R.P.C. § 4.1(a)(1). The Court also notes the Patent Office has promulgated the "USPTO Rules of Professional Conduct," which conforms to the Model Rules of Professional Conduct of the American Bar Association. *See* 37 C.F.R. § 11.100 *et seq*. The Patent Office's rules are virtually identical to the New Jersey Rules of Professional Conduct with respect to candor towards the tribunal and truthfulness in statements to others.

As a threshold argument, Merck argues that the jury's verdict prevents a finding of unclean hands. Merck Proposed Conclusions of Law ("COL") 46-54, ECF 407. According to Merck, the only unclean hands theory set forth in Gilead's interrogatory responses is predicated on Merck's derivation of the inventions claimed in the '499 and '712 Patents from Pharmasset's confidential disclosures. Since the jury found the claims of the '499 and '712 Patent were not invalid for lack of written description or lack of enablement, the priority date of the asserted claims is January 18, 2002. As a result, Merck argues that it could not have derived the invention from Pharmasset in 2004 because its invention was completely conceived by January 18, 2002.

The Court disagrees with Merck's view of Gilead's interrogatory responses and the jury's verdict. Gilead's interrogatory responses made clear that its unclean hands defense is based on the belief that Merck improperly derived information about Pharmasset's invention from Pharmasset's confidential disclosures. Gilead's Supp. Response to Interrogatories 9-10, ECF 218-2. These responses did not, as Merck argues, limit Gilead to a theory of unclean hands based on 35 U.S.C. § 102(f), also known "derivation," which states a person shall be entitled to a patent unless "he did not himself invent the subject matter." If Gilead's unclean hands disclosure was interpreted as only disclosing a theory of unclean hands based strictly on § 102(f), it would be entirely redundant of Gilead's § 102(f) invalidity defense. It would also allow Merck's misconduct in obtaining Pharmasset's confidential information during the 2004 phone call and subsequent litigation misconduct to go unchecked. Gilead's responses, instead, provide Gilead the ability to pursue an unclean hands defense covering circumstances where Merck improperly received information from Pharmasset. Thus, the jury's verdict, which did foreclose a § 102(f) invalidity defense, does not prevent Gilead from pursuing a defense of unclean hands.

Moving to Merck's alternative arguments, first, Merck argues that cases finding unclean hands have involved repeated and egregious misconduct involving an elaborate scheme to defraud. According to Merck, isolated instances of misconduct or conduct that is susceptible to innocuous explanations do not rise to the level of egregious misconduct. However, Merck's argument glosses over the serious and outrageous conduct in this case in which Merck engaged in litigation misconduct by presenting fabricated testimony and engaging in improper business practices. The

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cases Merck cites in support of its argument do not contain findings of lying, unethical business practices, and litigation misconduct and instead turn on the fact the Court did not have sufficient evidence to determine whether lying occurred. *See Excelled Sheepskin & Leather Coat Corp. v. Oregon Brewing Co.*, 2014 WL 3874193, at *10 (S.D.N.Y. Aug. 5, 2014) (finding defendant failed to present clear and convincing evidence that plaintiff's representations were inaccurate); *Top Grade Construction v. Fluoresco Lighting-Sign Maintenance*, 2012 WL 1122599, at *10 (N.D. Cal. Apr. 3, 2012) (denying summary judgment that plaintiff had unclean hands because defendant "presented no evidence to show that [p]laintiff intentionally misrepresented" information and there was a triable issue of fact as to whether plaintiff explanation for an inconsistent response is credible); *Lenz v. Universal Music Corp.*, 2010 U.S. Dist. LEXIS 16899, at *15-17 (N.D. Cal. Feb. 25, 2010) (no evidence any misstatements were made in bad faith); *Big Lots Stores, Inc. v. Jaredco, Inc.*, 182 F. Supp. 2d 644, 652 (S.D. Ohio 2002) (finding conduct was susceptible to more innocuous explanations because there was no evidence that a witness had lied or that counsel acted wrongfully and deceitfully); *In re Coordinated Pretrial Proceedings in Antibiotic Antitrust Actions (Pfizer, Inc. v. Int'l Rectifier Corp.)*, 538 F.2d 180, 195-196 (8th Cir. 1976) (any misstatements were an oversight because "the facts so concealed were basically supportive of [the concealing party's] contentions"); *Helene Curtis Indus. v. Sales Affiliates*, 121 F. Supp. 490, 510, 512 (S.D.N.Y. 1954) (holding unclean hands was not applicable because there was no evidence that the patentee had deliberately misrepresented or omitted information) .

Merck also attempts to downplay the seriousness of its misconduct by relying on post-*Therasense* cases that apply the egregious misconduct prong of inequitable conduct. Merck argues these cases find egregious misconduct in the presence of systematic and outrageous deception, or in other words, conduct that is more extreme than the conduct in this case. Merck Proposed COL ¶ 45, ECF 407 (citing *Apotex, Inc. v. UCB,* Inc., 970 F. Supp. 2d 1297, 1328 (S.D. Fla. 2013) (inventor's "overall pattern of misconduct" included "purposefully mislead[ing]" the Patent Office by misrepresenting invalidating prior art, withholding references, concealing detrimental test results, fabricating results for tests that were not conducted, and facilitating the submission of a misleading expert report), aff'd on other grounds, 763 F.3d 1354 (Fed. Cir. 2014);

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Intellect Wireless, Inc. v. HTC Corp., 732 F.3d 1339, 1342, 1343-44 (Fed. Cir. 2013) (inventor "filed multiple unmistakably false declarations during prosecution" to overcome prior art)). What Merck's argument fails to recognize is that the conduct in this case consists of systematic and outrageous deception in conjunction with unethical business practices and litigation misconduct. As discussed above, Merck violated its understanding with Pharmasset about who would receive structural information about PSI-6130. Compounding this problem, Merck attempted to minimize and conceal this behavior with Dr. Durette's fabricated testimony at his deposition and at trial. Even if the Court credits Merck's argument that it did not control the content of Dr. Durette's deposition testimony, the Court cannot ignore the fact that Merck never sought to correct the record until trial. And even then, Merck's witness continued to lie about what he knew and when he knew it.

Further relying on post-*Therasense* cases, Merck argues that misleading statements are not enough to rise to the level of egregious misconduct. Of course, the Court has found more than misleading statements. The Court has found that Merck engaged in improper business practices and litigation misconduct. That said, Merck's cases do not fully support its argument that misleading statements do not rise to the level of egregious misconduct; instead, those cases found that when it was ambiguous or not clear whether a statement was false, that uncertainty does not create egregious misconduct. *See Smith & Nephew, Inc. v. Interlace Med., Inc.*, 955 F. Supp. 2d 69 (D. Mass. 2013) (finding ambiguous misrepresentation was not egregious misconduct); *Network Signatures, Inc. v. State Farm Mut. Auto. Ins. Co.*, 2012 WL 2357307, at *7 (C.D. Cal. June 13, 2012) (not clear whether statement that delay in paying patent maintenance fee was unintentional was made to deceive the Patent Office), *rev'd on other grounds*, 731 F.3d 1239 (Fed. Cir. 2013); *Ohio Willow Wood Co. v. Alps S., LLC*, 735 F.3d 1333, 1339 (Fed. Cir. 2013) (denying summary judgment))

Second, Merck argues that its conduct is not improper because the law expressly allows it to file claims that cover a competitor's product. *See Kingsdown Med. Consultants, Ltd. v. Hollister Inc.*, 863 F.2d 867, 874 (Fed. Cir. 1988). In *Kingsdown*, the Federal Circuit stated:

> [T]here is nothing improper, illegal or inequitable in filing a patent

> application for the purpose of obtaining a right to exclude a known competitor's product from the market; nor is it in any manner improper to amend or insert claims intended to cover a competitor's product the applicant's attorney has learned about during the prosecution of a patent application. Any such amendment or insertion must comply with all statutes and regulations, of course, but, if it does, its genesis in the marketplace is simply irrelevant and cannot of itself evidence deceitful intent.

Id. (citing *State Indus., Inc. v. A.O. Smith Corp.*, 751 F.2d 1226, 1235 (Fed. Cir. 1985)). There are multiple problems with Merck's argument. First, Merck's argument relies on the assumption that it amended the claims to cover a competitor's product. But Dr. Durette testified that he amended the claims to cover the most important compounds in the Merck-Isis collaboration and not to cover Pharmasset's product. When pressed at trial, Dr. Durette refused to cleanly admit that he amended the claims to cover structures he saw in the Clark publication. Thus, Merck's argument fails to fit the evidence adduced during this case.

Even if that were not the case, the Court finds *Kingsdown*'s holding is premised entirely on the assumption that a patentee learns of a competitors' product through legal and ethical means. Here, Merck learned of PSI-6130, Pharmasset's crown jewel, during its due diligence of Pharmasset. This information was provided to Merck in a confidential setting to Merck employees who were purportedly firewalled from the prosecution of Merck's HCV patents. The Federal Circuit's holding in *Kingsdown* does not permit individuals to disregard firewalls and confidentiality agreements; holding otherwise, would bring the marketplace to a halt as companies would be weary to engage in due diligence lest a competitor uses that information to obtain patents.

Third, Merck claims Dr. Durette did not have an intent to deceive. Merck notes that "to meet the clear and convincing evidence standard, the specific intent to deceive must be the 'single most reasonable inference able to be drawn from the evidence.'' Merck Proposed COL ¶ 60, ECF 407 (quoting *Therasense*, 649 F.3d at 1290). According to Merck, Dr. Durette did not have an intent to deceive because he disclosed his conflict during the 2004 phone call and any further misstatements were simply the result of a lapse in memory. As support, Merck cites several cases where courts have refused to infer bad faith or intent to deceive from the fact of a misrepresentation, without more. Merck Proposed COL ¶¶ 64, 65, 66, 69, ECF 407 (citing *Outside the Box Innovations, LLC v. Travel Caddy, Inc.*, 695 F.3d 1285, 1294-95 (Fed. Cir. 2012); *Larson Mfg. Co. of S.D. v. Aluminart Prods. Ltd.*, 559 F.3d 1317, 1341 (Fed. Cir. 2009); *Excelled Sheepskin*, 2014 WL 3874193, at *10 (S.D.N.Y. Aug. 5, 2014); *Abbott Labs. v. Sandoz, Inc.*, 544 F.3d 1341, 1354-57 (Fed. Cir. 2008); *Eastman Kodak Co. v. Agfa-Gevaert N.V.*, 560 F. Supp. 2d 227, 301 (W.D.N.Y. 2008), judgment entered, 2008 WL 5115252 (W.D.N.Y. Dec. 4, 2008)). Merck also cites cases where courts have refused to infer intent to deceive from errors that could be due to memory lapses. Merck Proposed COL ¶ 67, ECF 407 (citing *BASF Corp. v. Aristo, Inc.*, 872 F. Supp. 2d 758, 779 (N.D. Ind. 2012); *United States v. Bailey*, 123 F.3d 1381, 1395 (11th Cir. 1997)). While Merck accurately conveys the holdings of the cases it cites, these cases are inapposite to the present facts, which involve substantially more than a "misrepresentation, without more" or "errors that *could* be due to memory lapses." As explained throughout this order, Merck's fabricated testimony was more than just an isolated incident, but happened repeatedly during Dr. Durette's deposition. At trial, Dr. Durette continued to be evasive and told a story that was not credible. Moreover, while perhaps a common and convenient post-fabrication excuse, a memory lapse does not explain Dr. Durette's confident and sanctimonious deposition testimony, nor does it explain Dr. Durette's sudden moments of purported clarity at trial, when for example, he magically recalled meeting with a supervisor prior to attending the 2004 phone call with Pharmasset. As such, the present facts are significantly more disturbing than those in any of the cases cited by Merck. The evidence in this case fully supports a finding of intent to deceive.

Fourth, Merck argues that Dr. Durette's conduct cannot be imputed to Merck. It argues that a non-litigant's misconduct cannot support unclean hands unless it is attributable to the litigant. Since Dr. Durette was no longer a Merck employee at the time of his deposition, was not under Merck's control, and was not a 30(b)(6) witness as to the subject of the 2004 call, Merck argues there is no basis to impute Dr. Durette's intent and conduct. Merck also argues it did not try to hide Dr. Durette's participation on the 2004 phone call, as it acknowledged that in its opening statement.

The Court disagrees with Merck and finds the evidence clearly supports imputing Dr.

Durette's conduct to Merck. Dr. Durette appeared at the deposition as Merck's designated 30(b)(6) corporate representative on issues related to the prosecution of the '499 Patent, including all reasons for amending any pending claim during prosecution. At the deposition, Dr. Durette was represented by Merck's outside counsel and leading up to the deposition, Dr. Durette met with Merck's outside and inside counsel for two full days of preparation, totaling 12 to 14 hours. Moreover, although Dr. Durette was outside the subpoena power of the Court, and Merck voluntarily brought Dr. Durette to trial on its behalf. Additionally, Merck presented Dr. Durette's testimony on direct examination to support its claim of patent validity. Finally, Merck's argument that it openly acknowledged Dr. Durette's participation in the 2004 phone call overlooks that in the very next sentence, its counsel told the jury that Dr. Durette appeared on the phone call because he did not know the compound that was going to be disclosed was within the scope of the Merck patent applications he was working on which turned out to be false. Thus, through Dr. Durette, Merck directed, advised, guided, and covered up misconduct and Merck argued on behalf of Dr. Durette throughout this proceeding. Accordingly, Dr. Durette's conduct may be imputed to Merck.

Moreover, the record amply supports the conclusion that while Dr. Durette was employed by Merck, his conduct was supervised by his managers. He testified that he had a pre-call meeting with his supervisor to discuss whether his HCV patent work would overlap Pharmasset's compound and during the 2004 call, he declared he would have to discuss the same issue with his supervisor. The only reasonable inference that can be drawn is that Dr. Durette continued to prosecute the '499 Patent under the direction of Merck.

Fifth, Merck argues that there is no immediate and necessary relation between the asserted claims and alleged misconduct. Merck claims that to prevail on its unclean hands defense, Gilead must show that the alleged misconduct (1) directly related to the claims Merck asserts in the present suit, and (2) as a result Gilead suffered injury. Merck Proposed COL ¶ 78 (citing *Hynix*, 897 F. Supp. 2d at 978). Merck's reliance on *Hynix* is not persuasive. *Hynix* did not establish a two-step test for showing the "immediate and necessary relation" component of unclean hands. Instead, the Court was reiterating the notion that misconduct must relate to the party asserting the

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defense and cannot be some general wrongdoing. *See id*. (citing *Dream Games of Ariz. Inc. v. PC Onsite*, 561 F.3d 983, 990 (9th Cir. 2009)). In *Dream Games*, the Ninth Circuit re-emphasized that under the longstanding principal of unclean hands, misconduct must relate to the party asserting the defense. *Id.*; *see also Republic Molding Corp. v. B.W. Photo Utilities*, 319 F.2d 347 (9th Cir. 1963) ("What is material is not that the plaintiff's hands are dirty, but that he dirtied them in acquiring the right he now asserts, or that the manner of dirtying renders inequitable the assertion of such rights against the defendant. As Professor Chafee suggests…, we should not by this doctrine create a rule comparable to that by which a careless motorist would be 'able to defend the subsequent personal injury suit by proving that the pedestrian had beaten his wife before leaving his home.'"). Here, as the Court has explained, the misconduct relates directly to Gilead as it involves Merck's misconduct with respect to Pharmasset and this litigation.

Furthermore, the thrust of Merck's argument is that Gilead did not suffer any harm because Merck did not obtain patent coverage that it would not have otherwise obtained. Merck Proposed COL ¶ 79, ECF 407. However, this argument would have the effect of imposing a non-existent materiality requirement onto unclean hands and further reveals the flaw in Merck's interpretation of the "immediate and necessary relation" component of unclean hands. While misconduct must relate to the asserted claims, which it does in this case, the misconduct does not have to be material. *See Therasense*, 649 F.3d at 1287 ("This court recognizes that the early unclean hands cases do not present any standard for materiality."). As a result, the Court finds Merck's argument is nothing more than an attempt to import a materiality requirement into unclean hands that would be inconsistent with Supreme Court authority.

Sixth, Merck argues that the '712 Patent is not unenforceable due to unclean hands. Merck claims that its in-house patent prosecutor, Mr. Jeffrey Bergman began working on the '712 Patent in 2011 and was responsible for narrowing the original claims. Since Mr. Bergman narrowed the amended claims and there is no evidence that Mr. Bergman engaged in misconduct, Merck argues there is no immediate and necessary relation between Dr. Durette's misconduct and the prosecution of the '712 Patent.

Contrary to Merck's argument, Merck and Dr. Durette's intentional litigation misconduct casts a darkness on this entire case that covers both patents-in-suit. Dr. Durette played a key role in the prosecution of both the ’499 and ’712 Patents. He was responsible for filing the application that eventually matured as the ’712 Patent and this application shares the same specification as the ’499 Patent. Although Merck cites several cases in support of its argument that the ’712 Patent is not affected by the misconduct, these cases deal with starkly different factual situations. In all of Merck’s cases, one party is trying to allege that misconduct related to a patent not-in-suit should give rise to unclean hands to an asserted patent. *See, e.g.*, *Advanced Magnetic Closures, Inc. v. Rome Fastener Corp.*, 2006 WL 3342655, at *1-2 (S.D.N.Y. Nov. 16, 2006) (rejecting unclean hands defense predicated on the wrongful assertion of other patents not involved in the litigation); *MedPointe Healthcare Inc. v. Hi-Tech Pharmacal Co.*, 380 F. Supp. 2d 457, 466 (D.N.J. 2005) (rejecting an assertion of unclean hands that at best involved plaintiff’s failure to disclose a prior ruling on a different, though related, patent, which was not the patent involved in the litigation); *Hoffman-La Roche, Inc. v. Promega Corp.*, 319 F. Supp. 2d 1011 (N.D. Cal. 2004) (rejecting unclean hands defense predicated on non-asserted patent). Here both the ’499 and ’712 Patents were asserted in this case; Merck and Dr. Durette’s litigation misconduct infected this entire case, covering both patents-in-suit. Moreover, it would be an odd result, to say the least, if Merck could engage in the substantial litigation misconduct exhibited in this case, yet face no penalty because the ’712 Patent was deemed uncontaminated.[5]

In sum, the Court concludes that Dr. Durette knowingly misled Pharmasset regarding his status as being within the firewall at the March 17, 2004, due diligence call. Merck approved this misconduct both before and after the March 17, 2004, call by initially assigning its HCV patent attorney to handle the Pharmasset due diligence work and thereafter, when Dr. Durette ceased his due diligence work on Pharmasset’s compound, directing him to remain active in prosecuting Merck’s overlapping HCV patent docket after Dr. Durette obtained the highly confidential Pharmasset PSI-6130 disclosure. Moreover, the Court concludes that Dr. Durette intentionally

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[5] The Court’s finding of improper business conduct related to the March 2004 call was not considered by the Court in determining whether unclean hands prevented enforcement of the ’712 Patent.

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fabricated testimony in this case and that Merck supported that bad faith conduct.

D. Balance of Equities

The last step of the unclean hands analysis is to balance the equities. “The Supreme Court has emphasized, however, that the doctrine of unclean hands ‘does not mean that courts must always permit a defendant wrongdoer to retain the profits of his wrongdoing merely because the plaintiff himself is possibly guilty of transgressing the law.’” *Northbay Wellness Grp., Inc. v. Beyries*, 789 F.3d 956, 960 (9th Cir. 2015) (quoting *Johnson v. Yellow Cab Transit Co.*, 321 U.S. 383, 387 (1944)). As the Ninth Circuit has explained:

> Unclean hands…does not stand as a defense that may be properly considered independent of the merits of the plaintiff's claim.... In the interests of right and justice the court should not automatically condone the defendant's infractions because the plaintiff is also blameworthy, thereby leaving two wrongs unremedied and increasing the injury to the public. Rather[,] the court must weigh the substance of the right asserted by plaintiff against the transgression which, it is contended, serves to foreclose that right. The relative extent of each party's wrong upon the other and upon the public should be taken into account, and an equitable balance struck. The ultimate decision is whether the deception actually caused by plaintiff as compared with the trading methods of the defendant warrant punishment of the plaintiff rather than of the defendant.

Republic Molding, 319 F.2d at 350.

Although there is no precise set of criterion for such balancing, courts have generally considered the weight of wrongdoing of one party against the wrongdoing of the other. For example, in *Hoffman-La Roche*, the Court considered the number of false statements made by the patentees in prosecuting their patents and found the balance of the equities did not favor the patentees. 319 F. Supp. 2d at 1015-16. In *Dunlop-McCullen v. Local 1-S*, 149 F. 3d 85, 92-93 (2d Cir. 1998), a case under the Labor-Management Reporting and Disclosure Act, the court denied defendant’s request to bar suit under the doctrine of unclean hands where the parties’ wrongful conduct was remarkably similar in quality and extent but where, on balance, the court found that defendant’s conduct was more significant so that the plaintiff was permitted to proceed with the suit. In *Northbay Wellness*, a bankruptcy case where a creditor sought by adversary proceeding to obtain a finding that a debt was nondischargeable based on theft, the Ninth Circuit was faced with

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balancing the seriousness of, on the one hand, an attorney's theft from his client of funds that led to his disbarment against, on the other hand, illegal marijuana sales by the other party. 789 F.3d at 960-61. Reversing the lower court, the Ninth Circuit held that the lower court had failed to conduct this balancing test and determined that unclean hands would not bar Northbay from its suit because, on balance, Northbay's board member, shared in its wrongdoing and his own culpability for theft of client funds was so egregious as to harm both Northbay and the public. *Id*.

In this case, Gilead is guilty of patent infringement. It admitted so much in response to Merck's motion for summary judgment, and on that basis, the Court granted summary judgment of infringement against Gilead. ECF 214. By contrast, Merck has engaged in business misconduct and litigation misconduct that the Court has found to be egregious.

As to Gilead's misconduct, it goes without saying that patent infringement is serious. However, in virtually every patent case where unclean hands is asserted, it comes on the heels of an infringement finding. *See Keystone*, 290 U.S. at 242; *Hazel-Atlas*, 322 U.S. 241-42; *Precision*, 324 U.S. at 814.

Merck raises a number of arguments to demonstrate that its conduct was less culpable than Gilead's. First, and foremost, Merck argues that Gilead's claim of unclean hands is weak. As described in detail above, the Court disagrees. The Court has determined that Merck engaged in a pervasive pattern of misconduct amply support by the evidence.

Merck further argues that there is no evidence that it intended to deceive Gilead or the Court. Again, the Court has found otherwise. From the evidence, it is clear to the Court that Merck's conduct during the Merck-Pharmasset discussions of allowing Dr. Durette to participate and assuring Merck, albeit falsely, that Dr. Durette was firewalled, its decision to allow Dr. Durette to continue to prosecute Merck's own HCV patent portfolio in violation of the firewall requirements and its own policy, its tainted judgment in amending the '499 claims 18 days after the Clark application published, its litigation misconduct including Dr. Durette's lying at his deposition, recanting that testimony at trial without proper prior notice to Gilead, and further untruthful testimony at trial all support the Court's conclusion that Merck did intend to deceive Gilead and the Court.

Next, Merck argues that the events in 2004 are irrelevant. Merck claims that Pharmasset knew that its PSI-6130 infringed Merck's patent applications. The Court has not made such a factual finding and on the record before it, cannot do so. Although there was evidence that Merck told Pharmasset that it did not have freedom to operate and that Jeremy Clark used the '499 Patent application to inform his lab work in developing PSI-6130, the evidence further shows that Pharmasset rejected Merck's accusations and that it reviewed the '499 application in order to expressly stay clear of infringement. On this record, the Court does not find the 2004 events irrelevant.

Merck further argues that it did not engage in misconduct before the PTO. While true, good behavior in one setting does not absolve Merck's misconduct in this setting. Additionally, unlike the inequitable conduct defense, misconduct is not limited to the PTO forum. *Therasense*, 649 F.3d at 1287.

Merck argues that Gilead was not harmed by its conduct. But this argument does not align with case law. The balancing of the equities analysis is not limited to the private harm caused by the misconduct. To say otherwise would impose a materiality requirement where there is none. *Id*. Rather, the focus is on the transgressions of both parties, to make sure that two wrongs are not left unpunished against the public interest. Even assuming that Merck is correct on this point, there was a significant public harm regarding false testimony and improper business conduct that permeated this suit.

Merck also argues that barring it from suit against Gilead is far too severe a penalty for its conduct. The Court acknowledges that the jury's damages award demonstrates the significance of the rights at risk. Taking that into account, however, it is the Court's determination that, on balance, Merck's persistent misconduct involving repeated fabricated testimony and improper business conduct outweigh its right to maintain this suit against Gilead.

As oft repeated, Learned Hand stated:

> The doctrine is confessedly derived from the unwillingness of a court, originally and still nominally one of conscience, to give its peculiar relief to a suitor who in the very controversy has so conducted himself as to shock the moral sensibilities of the judge. It has nothing to do with the

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> rights or liabilities of the parties; indeed the defendant who invokes it need not be damaged, and the court may even raise it sua sponte.

Saudi Basic Indus. Corp. v. ExxonMobil Corp., 401 F.Supp.2d 383, 392-93 (D.N.J. 2005) (quoting *Gaudiosi v. Mellon*, 269 F.2d 873, 882 (3rd Cir. 1959)). For the foregoing reasons, a balance of the equities favors Gilead, and thus, the Court concludes that Gilead has proven its defense of unclean hands by clear and convincing evidence.

VI. CONCLUSION

Candor and honesty define the contours of the legal system. When a company allows and supports its own attorney to violate these principles, it shares the consequences of those actions. Here, Merck's patent attorney, responsible for prosecuting the patents-in-suit, was dishonest and duplicitous in his actions with Pharmasset, with Gilead and with this Court, thus crossing the line to egregious misconduct. Merck is guilty of unclear hands and forfeits its right to prosecute this action against Gilead.

VII. ORDER

For the foregoing reasons, IT IS HEREBY ORDERED that Merck is barred from asserting the '499 and '712 Patents against Gilead and Merck shall take nothing by this suit.

IT IS SO ORDERED.

Dated: June 6, 2016

Beth Labson Freeman

BETH LABSON FREEMAN
United States District Judge

United States District Court
Northern District of California

UNITED STATES DISTRICT COURT

NORTHERN DISTRICT OF CALIFORNIA

SAN JOSE DIVISION

GILEAD SCIENCES, INC.,
Plaintiff,
v.
MERCK & CO, INC., et al.,
Defendants.

Case No. 13-cv-04057-BLF

ORDER DENYING GILEAD'S MOTION FOR JUDGMENT AS A MATTER OF LAW AND ENTERING FINAL JUDGMENT

On consideration of Gilead's Motion for Judgment as a Matter of Law (ECF No. 432), it is hereby ORDERED that Gilead's Motion is DENIED as moot, because the Court has previously held that unclean hands "renders Merck's '499 and '712 Patents unenforceable against Gilead." ECF No. 422; *cf*. Wright & Miller, 13 B Fed. Prac. & Proc. Juris. § 3533 (3d ed.) ("When a court grants full relief on one ground, it may refer to an alternative ground that might support the same relief as moot."). This resolves all pending issues except for a collateral matter—the amount of attorney's fees that should be awarded to Gilead. Final judgment is hereby entered.

IT IS SO ORDERED.

Dated: August 16, 2016

Beth Labson Freeman
BETH LABSON FREEMAN
United States District Judge

US007105499B2

(12) **United States Patent**
Carroll et al.

(10) **Patent No.: US 7,105,499 B2**
(45) **Date of Patent: Sep. 12, 2006**

(54) **NUCLEOSIDE DERIVATIVES AS INHIBITORS OF RNA-DEPENDENT RNA VIRAL POLYMERASE**

(75) Inventors: **Steven S. Carroll**, Yardley, PA (US); **David B. Olsen**, Lansdale, PA (US); **Malcolm MacCoss**, Freehold, NJ (US); **Balkrishen Bhat**, Carlsbad, CA (US); **Phillip Dan Cook**, Fallbrook, CA (US); **Anne B. Eldrup**, Encinitas, CA (US); **Thazha P. Prakash**, Carlsbad, CA (US)

(73) Assignees: **Merck & Co., Inc.**, Rahway, NJ (US); **Isis Pharmaceuticals, Inc.**, Carlsbad, CA (US)

(*) Notice: Subject to any disclaimer, the term of this patent is extended or adjusted under 35 U.S.C. 154(b) by 335 days.

(21) Appl. No.: **10/250,873**

(22) PCT Filed: **Jan. 18, 2002**

(86) PCT No.: **PCT/US02/01531**

§ 371 (c)(1), (2), (4) Date: **Jan. 16, 2004**

(87) PCT Pub. No.: **WO02/057425**

PCT Pub. Date: **Jul. 25, 2002**

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Related U.S. Application Data

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(51) **Int. Cl.**

A61K 31/7052	(2006.01)
A61K 31/7042	(2006.01)
A61K 31/7068	(2006.01)
C07H 19/04	(2006.01)
C07H 19/06	(2006.01)

(52) **U.S. Cl.** **514/49**; 514/43; 514/45; 514/42; 514/50; 514/52; 514/46; 536/28.4; 536/28.5; 536/26.12

(58) **Field of Classification Search** 514/49, 514/43, 45, 42, 50, 52, 46; 536/28.4, 28.5, 536/26.12, 26.13
See application file for complete search history.

(56) **References Cited**

U.S. PATENT DOCUMENTS

3,480,613	A		11/1969	Walton
6,348,587	B1	*	2/2002	Schinazi et al. 536/25.3
6,784,161	B1	*	8/2004	Ismaili et al. 514/43
2002/0019363	A1		2/2002	Ismaili et al.
2002/0055483	A1		5/2002	Watanabe et al.
2002/0198171	A1		12/2002	Schinazi et al.
2003/0050229	A1		3/2003	Sommadossi et al.
2003/0060400	A1		3/2003	LaColla et al.

FOREIGN PATENT DOCUMENTS

FR	1.581.628		9/1969
GB	1209 654		10/1970
WO	WO 93/18051		9/1993
WO	WO 99/43691		9/1999
WO	WO 01/32153		5/2001
WO	WO 01/60315		8/2001
WO	WO 01/79246		10/2001
WO	WO 01/90121		11/2001
WO	WO 01/92282		12/2001
WO	WO 02/18404		3/2002
WO	WO 02/048165	A3	6/2002
WO	WO 02/048165	A2	6/2002
WO	WO 02/069903	A3	9/2002
WO	WO 02/069903	A2	9/2002
WO	WO 02/094289	A1	11/2002
WO	WO 02/100354	A3	12/2002
WO	WO 02/100354	A2	12/2002
WO	WO 02/100415	A2	12/2002
WO	WO 03/000200	A2	1/2003
WO	WO 03/000713	A1	1/2003
WO	WO 03/015798	A1	2/2003
WO	WO 03/026589	A2	4/2003
WO	WO 03/026675	A1	4/2003
WO	WO 03/051881		6/2003
WO	WO 03/051896		6/2003
WO	WO 03/051897		6/2003
WO	WO 03/051898		6/2003
WO	WO 03/051899		6/2003
WO	WO 03/062255		7/2003

OTHER PUBLICATIONS

Walton, E. et al., "Branched–Chain Sugar Nucleosides. V. Synthesis and Antiviral Properties of Several Branched–Chain Sugar Nucleosides", J. Med. Chem., vol. 12, pp. 306–309 (1969).

(Continued)

Primary Examiner—Elvis O. Price
Assistant Examiner—Michael C. Henry
(74) *Attorney, Agent, or Firm*—Philippe L. Durette; Melvin Winokur

(57) **ABSTRACT**

The present invention provides nucleoside derivatives which are inhibitors of RNA-dependent RNA viral polymerase. These compounds are inhibitors of RNA-dependent RNA viral replication and are useful for the treatment of RNA-dependent RNA viral infection. They are particularly useful as inhibitors of hepatitis C virus (HCV) NS5B polymerase, as inhibitors of HCV replication, and/or for the treatment of hepatitis C infection. The invention also describes pharmaceutical compositions containing such nucleoside derivatives alone or in combination with other agents active against RNA-dependent RNA viral infection, in particular HCV infection. Also disclosed are methods of inhibiting RNA-dependent RNA polymerase, inhibiting RNA-dependent RNA viral replication, and/or treating RNA-dependent RNA viral infection with the nucleoside derivatives of the present invention.

2 Claims, No Drawings

OTHER PUBLICATIONS

Matsuda, A. et al., "Radical Deoxygenation of Tert–Alcohols In 2'–Branched–Chain Sugar Pyrimidine Nucleosides: Synthesis and Antileukemic Activity of 2'–Deoxy–2' (S)–Methylcytidine", Chem. Pharm. Bull., vol. 35, pp. 3967–3970 (1987).

Matsuda, A. et al., Alkyl Addition Reaction of Pyrimidine 2'–Ketonucleosides: Synthesis of 2'–Branched–Chain Sugar Pyrimidine Nucleosides (Nucleosides and Nucleotides. LXXXI). Chem. Pharm. Bull., vol. 36, pp. 945–953 (1988).

Murai, Y. et al., "A Synthesis and an X–Ray Analysis of 2'–C–, 3'–C– and 5'–C–Methylsangivamycins", Heterocycles, vol. 33, pp. 391–404 (1992).

Limori, T. et al., "2'–C–, 3'–C– and 5'–C–Methylsangivamycins: Conformational Lock with the Methyl Group", Tetrahedron Letters, vol. 3, No. 49, pp. 7273–7276 (1991).

Wolfe, M. et al., "A Consise Synthesis of 2'–C–Methylribonucleosides", Tetrahedron Letters, vol. 36, No. 42, pp. 7611–7614 (1995).

Harry–O'kuru, R. et al., "A Short, Flexible Route toward 2'–C–Branched Ribonucleosides", J. Org. Chem., vol. 62, pp. 1754–1759 (1997).

Gallo, M. et al., "Synthesis of 2'–Modified Nucleotides", Molecules, vol. 5, pp. 727–729 (2000).

Beigelman, L. et al., "New Synthesis of 2'–C–Methylnucleosides Starting From D–Glucose and D–Ribose", Carbohydrate Research, vol. 166, pp. 219–232 (1987).

Anzai, K. et al, "A New Antibiotic, Tubercidin", The Journal of Antibiotics., vol. X, No. 5, pp. 201–205 (1957).

Tolman, R. et al., "Pyrrolopyrimidine Nucleosides. III. The Total Synthesis of Toyocamycin, Sangivamycin, Tubercidin, and Related Derivatives", Journal of the American Chemical Society, vol. 91, No. 8, pp. 2102–2108 (1969).

Tolman, R. et al., "Pyrrolo[2,3–d]pyrimidine Nucleoside Antibiotics. Total Synthesis and Structure of Toyacamycin, Unamycin B, Vengicide, Antibiotic E–212, and Sangivamycin (BA–90212)", Journal of the American Chemical Society, vol. 90, No. 2, pp. 24–526 (1968).

Siddiqi, S. et al., "Search for New Purine– and Ribose–Modified Adenosine Analogues as Selective Agonists and Antagonists at Adenosine Receptors", J. Med. Chem., vol. 38, pp. 1174–1188 (1995).

Franchetti, P. et al., "2'–Methyl Analogues of Selective Adenosine Receptor Agonists: Synthesis and Binding Studies", J. Med. Chem., vol. 41, pp. 1708–1715 (1988).

Kalinichenko, E. et al., "Substrate Specificity of Adenosine Deminase. Role of Methyl Groups at the 2'–3'–, and 5'–Carbon Atoms of Adenosine", Bioorganicheskaya Khimiya, vol. 14, No. 9, pp. 1157–1161 (1988).

Wolf, et al., "New 2'–C–Branched–Chain Sugar Nucleoside Analogs with Potential Antiviral or Antitumor Activity", Synthesis, pp. 773–778 (Aug. 1992).

* cited by examiner

NUCLEOSIDE DERIVATIVES AS INHIBITORS OF RNA-DEPENDENT RNA VIRAL POLYMERASE

CROSS REFERENCE TO RELATED APPLICATIONS

This application is the National Stage of International Application No. PCT/US2002/01531, filed 9 Jul. 2003, which claims the benefit under 35 U.S.C. 119(e) of U.S. provisional application No. 60/344,528, filed on Oct. 25, 2001; provisional application No. 60/299,320, filed on Jun. 19, 2001; provisional application No. 60/282,069, filed on Apr. 6, 2001; and provisional application No. 60/263,313, filed on Jan. 22, 2001; the contents of each of which are hereby incorporated by reference.

FIELD OF THE INVENTION

The present invention provides nucleoside derivatives which are inhibitors of RNA-dependent RNA viral polymerase. These compounds are inhibitors of RNA-dependent RNA viral replication and are useful for the treatment of RNA-dependent RNA viral infection. They are particularly useful as inhibitors of hepatitis C virus (HCV) NS5B polymerase, as inhibitors of HCV replication, and for the treatment of hepatitis C infection.

BACKGROUND OF THE INVENTION

Hepatitis C virus (HCV) infection is a major health problem that leads to chronic liver disease, such as cirrhosis and hepatocellular carcinoma, in a substantial number of infected individuals, estimated to be 2–15% of the world's population. There are an estimated 4.5 million infected people in the United States alone, according to the U.S. Center for Disease Control. According to the World Health Organization, there are more than 200 million infected individuals worldwide, with at least 3 to 4 million people being infected each year. Once infected, about 20% of people clear the virus, but the rest harbor HCV the rest of their lives. Ten to twenty percent of chronically infected individuals eventually develop liver-destroying cirrhosis or cancer. The viral disease is transmitted parenterally by contaminated blood and blood products, contaminated needles, or sexually and vertically from infected mothers or carrier mothers to their off-spring. Current treatments for HCV infection, which are restricted to immunotherapy with recombinant interferon-α alone or in combination with the nucleoside analog ribavirin, are of limited clinical benefit. Moreover, there is no established vaccine for HCV. Consequently, there is an urgent need for improved therapeutic agents that effectively combat chronic HCV infection. The state of the art in the treatment of HCV infection has been reviewed, and reference is made to the following publications: B. Dymock, et al., "Novel approaches to the treatment of hepatitis C virus infection," *Antiviral Chemistry & Chemotherap.* 11: 79–96 (2000); H. Rosen, et al., "Hepatitis C virus: current understanding and prospects for future therapies," *Molecular Medicine Today*, 5: 393–399 (1999); D. Moradpour, et al., "Current and evolving therapies for hepatitis C," *European J. Gastroenterol. Hepatol.*, 11: 1189–1202 (1999); R. Bartenschlager, "Candidate Targets for Hepatitis C Virus-Specific Antiviral Therapy," *Intervirology*, 40: 378–393 (1997); G. M. Lauer and B. D. Walker, "Hepatitis C Virus Infection," *N. Engl. J. Med.*, 345: 41–52 (2001); B. W. Dymock, "Emerging therapies for hepatitis C virus infection," *Emerging Drugs*, 6: 13–42 (2001); and C. Crabb, "Hard-Won Advances Spark Excitement about Hepatitis C," *Science*: 506–507 (2001); the contents of all of which are incorporated by reference herein in their entirety.

Different approaches to HCV therapy have been taken, which include the inhibition of viral serine proteinase (NS3 protease), helicase, and RNA-dependent RNA polymerase (NS5B), and the development of a vaccine.

The HCV virion is an enveloped positive-strand RNA virus with a single oligoribonucleotide genomic sequence of about 9600 bases which encodes a polyprotein of about 3,010 amino acids. The protein products of the HCV gene consist of the structural proteins C, E1, and E2, and the non-structural proteins NS2, NS3, NS4A and NS4B, and NS5A and NS5SB. The nonstructural (NS) proteins are believed to provide the catalytic machinery for viral replication. The NS3 protease releases NS5B, the RNA-dependent RNA polymerase from the polyprotein chain. HCV NS5B polymerase is required for the synthesis of a double-stranded RNA from a single-stranded viral RNA that serves as a template in the replication cycle of HCV. NS5B polymerase is therefore considered to be an essential component in the HCV replication complex [see K. Ishi, et al., "Expression of Hepatitis C Virus NS5B Protein: Characterization of Its RNA Polymerase Activity and RNA Binding," *Hepatology*, 29: 1227–1235 (1999) and V. Lohmann, et al., "Biochemical and Kinetic Analyses of NS5B RNA-Dependent RNA Polymerase of the Hepatitis C Virus," *Virology*, 249: 108–118 (1998)]. Inhibition of HCV NS5B polymerase prevents formation of the double-stranded HCV RNA and therefore constitutes an attractive approach to the development of HCV-specific antiviral therapies.

It has now been found that nucleoside compounds of the present invention and certain derivatives thereof are potent inhibitors of RNA-dependent RNA viral replication and in particular HCV replication. The 5'-triphosphate derivatives of the nucleoside compounds are inhibitors of RNA-dependent RNA viral polymerase and in particular HCV NS5B polymerase. The instant nucleoside compounds and derivatives thereof are useful to treat RNA-dependent RNA viral infection and in particular HCV infection.

It is therefore an object of the present invention to provide nucleoside compounds and certain derivatives thereof which are useful as inhibitors of RNA-dependent RNA viral polymerase and in particular as inhibitors of HCV NS5B polymerase.

It is another object of the present invention to provide nucleoside derivatives which are useful as inhibitors of the replication of an RNA-dependent RNA virus and in particular as inhibitors of the replication of hepatitis C virus.

It is another object of the present invention to provide nucleoside compounds and certain derivatives which are useful in the treatment of RNA-dependent RNA viral infection and in particular in the treatment of HCV infection.

It is another object of the present invention to provide pharmaceutical compositions comprising the novel compounds of the present invention in association with a pharmaceutically acceptable carrier.

It is another object of the present invention to provide pharmaceutical compositions comprising the nucleoside compounds and derivatives thereof for use as inhibitors of RNA-dependent RNA viral polymerase and in particular as inhibitors of HCV NS5B polymerase.

It is another object of the present invention to provide pharmaceutical compositions comprising the nucleoside compounds and derivatives thereof for use as inhibitors of RNA-dependent RNA viral replication and in particular as inhibitors of HCV replication.

It is another object of the present invention to provide pharmaceutical compositions comprising the nucleoside compounds and derivatives thereof for use in the treatment of RNA-dependent RNA viral infection and in particular in the treatment of HCV infection.

It is another object of the present invention to provide pharmaceutical compositions comprising the nucleoside compounds and derivatives thereof in combination with other agents active against an RNA-dependent RNA virus and in particular against HCV.

It is another object of the present invention to provide methods for the inhibition of RNA-dependent RNA viral polymerase and in particular for the inhibition of HCV NS5B polymerase.

It is another object of the present invention to provide methods for the inhibition of RNA-dependent RNA viral replication and in particular for the inhibition of HCV replication.

It is another object of the present invention to provide methods for the treatment of RNA-dependent RNA viral infection and in particular for the treatment of HCV infection.

It is another object of the present invention to provide methods for the treatment of RNA-dependent RNA viral infection in combination with other agents active against RNA-dependent RNA virus and in particular for the treatment of HCV infection in combination with other agents active against HCV.

It is another object of the present invention to provide nucleoside compounds and certain derivatives thereof and their pharmaceutical compositions for use as a medicament for the inhibition of RNA-dependent RNA viral replication and/or the treatment of RNA-dependent RNA viral infection and in particular for the inhibition of HCV replication and/or the treatment of HCV infection.

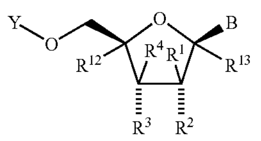
It is another object of the present invention to provide for the use of the nucleoside compounds and certain derivatives thereof of the present invention and their pharmaceutical compositions for the manufacture of a medicament for the inhibition of RNA-dependent RNA viral replication and/or the treatment of RNA-dependent RNA viral infection and in particular for the inhibition of HCV replication and/or the treatment of HCV infection.

These and other objects will become readily apparent from the detailed description which follows.

SUMMARY OF THE INVENTION

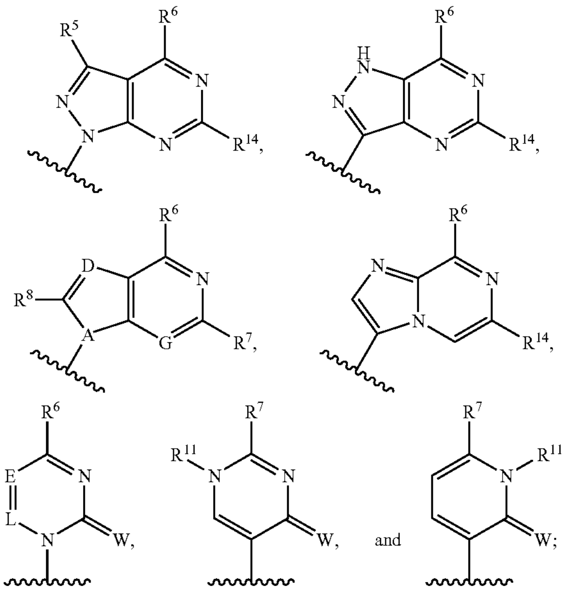
The present invention provides a method for inhibiting RNA-dependent RNA viral polymerase, a method for inhibiting RNA-dependent RNA viral replication, and/or a method for treating RNA-dependent viral infection in a mammal in need thereof, comprising administering to the mammal a therapeutically effective amount of a compound of structural formula I which is of the stereochemical configuration:

(I)



or a pharmaceutically acceptable salt thereof;

wherein B is selected from the group consisting of



A, G, and L are each independently CH or N;

D is N, CH, C—CN, C—NO_2, C—C_{1-13} alkyl, C—$NHCONH_2$, C—$CONR^{11}R^{11}$, C—$CSNR^{11}R^{11}$, C—$COOR^{11}$, C—C(=NH)NH_2, C-hydroxy, C—C_{1-3} alkoxy, C-amino, C—C_{1-4} alkylamino, C-di(C_{1-4} alkyl)amino, C-halogen, C-(1,3-oxazol-2-yl), C-(1,3-thiazol-2-yl), or C-(imidazol-2-yl); wherein alkyl is unsubstituted or substituted with one to three groups independently selected from halogen, amino, hydroxy, carboxy, and C_{1-3} alkoxy;

E is N or CR^5;

W is O or S;

Y is H, C_{1-10} alkylcarbonyl, $P_3O_9H_4$, $P_2O_6H_3$, or P(O)R^9R^{10};

R^1 is hydrogen, C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms and one of R^2 and R^3 is hydroxy or C_{1-4} alkoxy and the other of R^2 and R^3 is selected from the group consisting of

hydrogen,
hydroxy,
halogen,
C_{1-4} alkyl, optionally substituted with 1 to 3 fluorine atoms,
C_{1-10} alkoxy, optionally substituted with C_{1-3} alkoxy or 1 to 3 fluorine atoms,
C_{2-6} alkenyloxy,
C_{1-4} alkylthio,
C_{1-8} alkylcarbonyloxy,
aryloxycarbonyl,
azido,
amino,
C_{1-4} alkylamino, and
di(C_{1-4} alkyl)amino; or

R^2 is hydrogen, C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms and one of R^1 and R^3 is hydroxy or C_{1-4} alkoxy and the other of R^1 and R^3 is selected from the group consisting of hydrogen,

hydroxy,

halogen,

C_{1-4} alkyl, optionally substituted with 1 to 3 fluorine atoms,

C_{1-10} alkoxy, optionally substituted with hydroxy, C_{1-3} alkoxy, carboxy, or 1 to 3 fluorine atoms,

C_{2-6} alkenyloxy,

C_{1-4} alkylthio,

C_{1-8} alkylcarbonyloxy,

aryloxycarbonyl,

azido,

amino,

C_{1-4} alkylamino, and

di(C_{1-4} alkyl)amino; or

R^1 and R^2 together with the carbon atom to which they are attached form a 3- to 6-membered saturated monocyclic ring system optionally containing a heteroatom selected from O, S, and NC_{0-4} alkyl;

R^4 and R^6 are each independently H, OH, SH, NH_2, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, C_{3-6} cycloalkylamino, halogen, C_{1-4} alkyl, C_{1-4} alkoxy, or CF_3;

R^5 is H, C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-4} alkylamino, CF_3, or halogen;

R^{14} is H, CF_3, C_{1-4} alkyl, amino, C_{1-4} alkylamino, C_{3-6} cycloalkylamino, or di(C_{1-4} alkyl)amino;

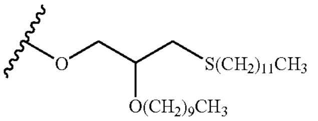
R^7 is hydrogen, amino, C_{1-4} alkylamino, C_{3-6} cycloalkylamino, or di(C_{1-4} alkyl)amino;

each R^{11} is independently H or C_{1-6} alkyl;

R^8 is H, halogen, CN, carboxy, C_{1-4} alkyloxycarbonyl, N_3, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, hydroxy, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} alkylsulfonyl, or (C_{1-4} alkyl)$_{0-2}$ aminomethyl;

R^{12} and R^{13} are each independently hydrogen, methyl, hydroxymethyl, or fluoromethyl; and

R^9 and R^{10} are each independently hydroxy, $OCH_2CH_2SC({=}O)C_{1-4}$ alkyl, $OCH_2O(C{=}O)OC_{1-4}$ alkyl, $NHCEMeCO_2Me$, $OCH(C_{1-4}\text{ alkyl})O(C{=}O)C_{1-4}$ alkyl,



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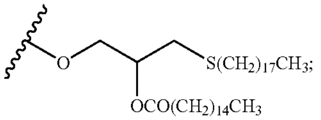
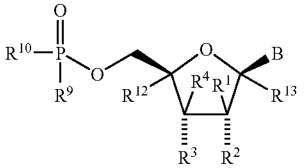


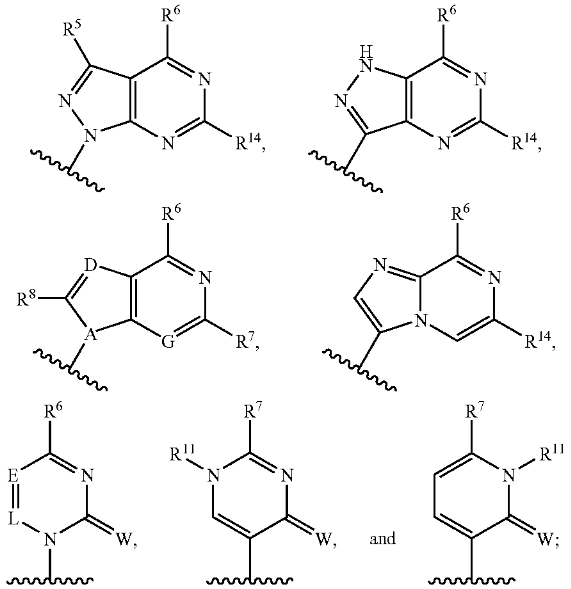
with the provisos that (a) when R^1 is hydrogen, one of R^3 and R^4 is hydrogen, and R^2 is fluoro, then the other of R^3 and R^4 is not hydrogen, halogen, azido, trifluoromethyl, C_{1-4} alkyl, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, or C_{1-10} alkoxy; (b) when R^1 is hydrogen, one of R^3 and R^4 is hydrogen, and R^2 is halogen, hydroxy, C_{1-6} alkoxy, or C_{2-6} alkenyloxy, then the other of R^3 and R^4 is not hydrogen, fluoro, or azido; and (c) when R^1 and R^3 are hydrogen and R^2 is hydroxy, then R^4 is not hydroxy.

The present invention also provides novel compounds of structural formula IV of the indicated stereochemical configuration which are useful as inhibitors of RNA-dependent RNA viral polymerase. The compounds of formula IV are also inhibitors of RNA-dependent RNA viral replication and are useful for the treatment of RNA-dependent RNA viral infection:

(IV)



wherein B is selected from the group consisting of



A, G, and L are each independently CH or N;

D is N, CH, C—CN, C—NO_2, C—C_{1-3} alkyl, C—$NHCONH_2$, C—$CONR^{11}R^{11}$, C—$CSNR^{11}R^{11}$, C—$COOR^{11}$, C—C(=NH)NH_2, C-hydroxy, C—C_{1-3} alkoxy, C-amino, C—C_{1-4} alkylamino, C-di(C_{1-4} alkyl)amino, C-halogen, C-(1,3-oxazol-2-yl), C-(1,3-thiazol-2-yl), or C-(imidazol 2-yl); wherein alkyl is unsubstituted or substituted with one to three groups independently selected from halogen, amino, hydroxy, carboxy, and C_{1-3} alkoxy;

E is N or CR^5;

W is O or S;

R^1 is hydrogen, C_{2-4} alkenyl, C_{2-4}-alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms and one of R^2 and R^3 is hydroxy or C_{1-4} alkoxy and the other of R^2 and R^3 is selected from the group consisting of

hydrogen,

hydroxy,

halogen,

C_{1-4} alkyl, optionally substituted with 1 to 3 fluorine atoms,

C_{1-10} alkoxy, optionally substituted with C_{1-3} alkoxy or 1 to 3 fluorine atoms,

C_{2-6} alkenyloxy,

C_{1-4} alkylthio,

C_{1-8} alkylcarbonyloxy,

aryloxycarbonyl,

azido,

amino,

C_{1-4} alkylamino, and

di(C_{1-4} alkyl)amino; or

R^2 is hydrogen, C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms and one of R^1 and R^3 is hydroxy or C_{1-4} alkoxy and the other of R^1 and R^3 is selected from the group consisting of

hydrogen,

hydroxy,

halogen,

C_{1-4} alkyl, optionally substituted with 1 to 3 fluorine atoms,

C_{1-10} alkoxy, optionally substituted with hydroxy, C_{1-3} alkoxy, carboxy, or 1 to 3 fluorine atoms,

C_{2-6} alkenyloxy,

C_{1-4} alkylthio,

C_{1-8} alkylcarbonyloxy,

aryloxycarbonyl,

azido,

amino,

C_{1-4} alkylamino, and

di(C_{1-4} alkyl)amino; or

R^1 and R^2 together with the carbon atom to which they are attached form a 3- to 6-membered saturated monocyclic ring system optionally containing a heteroatom selected from O, S, and NC_{0-4} alkyl;

each R^4 is independently H, OH, SH, NH_2, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, C_{3-6} cycloalkylamino, halogen, C_{1-4} alkyl, C_{1-4} alkoxy, or CF_3;

R^4 and R^6 are each independently H, OH, SH, NH_2, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, C_{3-6} cycloalkylamino, halogen, C_{1-4} alkyl, C_{1-4} alkoxy, or CF_3;

R^5 is H, C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-4} alkylamino, CF_3, or halogen;

R^{14} is H, CF_3, C_{1-4} alkyl, amino, C_{1-4} alkylamino, C_{3-6} cycloalkylamino, or di(C_{1-4} alkyl)amino;

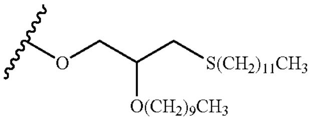
R^7 is hydrogen, amino, C_{1-4} alkylamino, C_{3-6} cycloalkylamino, or di(C_{1-4} alkyl)amino;

each R^{11} is independently H or C_{1-6} alkyl;

R^8 is H, halogen, CN, carboxy, C_{1-4} alkyloxycarbonyl, N_3, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, hydroxy, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} alkylsulfonyl, or (C_{1-4} alkyl)$_{0-2}$ aminomethyl;

R^{12} and R^{13} are each independently hydrogen, methyl, hydroxymethyl, or fluoromethyl; and

R^9 and R^{10} are each independently hydroxy, $OCH_2CH_2SC({=}O)C_{1-4}$ alkyl, $OCH_2O(C{=}O)OC_{1-4}$ alkyl, $NHCHMeCO_2Me$, $OCH(C_{1-4}$ alkyl$)O(C{=}O)C_{1-4}$ alkyl,



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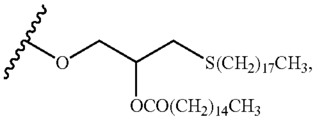
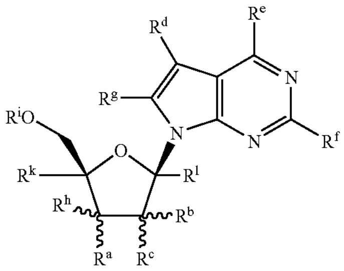


provided that at least one of R^9 and R^{10} is not hydroxy.

The present invention further provides novel compounds of structural formula XII of the indicated stereochemical configuration which are useful as inhibitors of RNA-dependent RNA viral polymerase and in particular of HCV NS5B polymerase:

(XII)



wherein R^a and R^h are each independently selected from the group consisting of hydrogen, cyano, azido, halogen, hydroxy, mercapto, amino, C_{1-4} alkoxy, C_{2-4} alkenyl, C_{2-4} alkynyl, and C_{1-4} alkyl, wherein alkyl is unsubstituted or substituted with hydroxy, amino, C_{1-4} alkoxy, C_{1-4} alkylthio, or one to three fluorine atoms;

R^b is C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl, wherein alkyl is unsubstituted or substituted with hydroxy, amino, C_{1-4} alkoxy, C_{1-4} alkylthio, or one to three fluorine atoms;

R^c is hydrogen, fluorine, hydroxy, mercapto, C_{1-4} alkoxy, or C_{1-4} alkyl; or R^b and

R^c together with the carbon atom to which they are attached form a 3- to 6-membered saturated monocyclic ring system optionally containing a heteroatom selected from O, S, and NC_{0-4} alkyl;

R^d is hydrogen, cyano, nitro, C_{1-3} alkyl, $NHCONH_2$, CONRjRj, CSNRjRj, COORj, $C({=}NH)NH_2$, hydroxy, C_{1-3} alkoxy, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, halogen, (1,3-oxazol-2-yl), (1,3-thiazol-2-yl), or (imidazol-2-yl); wherein alkyl is unsubstituted or substituted with one to three groups independently selected from halogen, amino, hydroxy, carboxy, and C_{1-3} alkoxy;

R^e and R^f are each independently hydrogen, hydroxy, halogen, C_{1-4} alkoxy, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, C_{3-6} cycloalkylamino, di(C_{3-6} cycloalkyl)amino, or C_{4-6} cycloheteroalkyl, unsubstituted or substituted with one to two groups independently selected from halogen, hydroxy, amino, C_{1-4} alkyl, and C_{1-4} alkoxy;

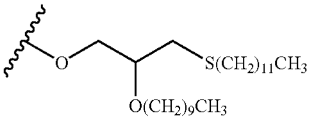
R^g is hydrogen, C_{1-4} alkyl, C_{2-4} alkynyl, halogen, cyano, carboxy, C_{1-4} alkyloxycarbonyl, azido, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, hydroxy, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} alkylsulfonyl, (C_{1-4} alkyl)$_{0-2}$ aminomethyl, or C_{4-6} cycloheteroalkyl, unsubstituted or substituted with one to two groups independently selected from halogen, hydroxy, amino, C_{1-4} alkyl, and C_{1-4} alkoxy;

R^i is hydrogen, C_{1-10} alkylcarbonyl, $P_3O_9H_4$, $P_2O_6H_3$, or $P(O)R^mR^n$;

each R^i is independently hydrogen or C_{1-6} alkyl;

R^k and R^l are each independently hydrogen, methyl, hydroxymethyl, or fluoromethyl; and

R^m and R^n are each independently hydroxy, $OCH_2CH_2SC({=}O)C_{1-4}$ alkyl, $OCH_2O(C{=}O)OC_{1-4}$ alkyl, $NHCHMeCO_2Me$, $OCH(C_{1-4}$ alkyl$)O(C{=}O)C_{1-4}$ alkyl,



or

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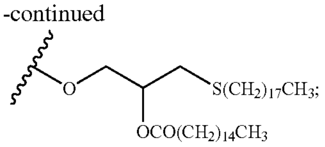


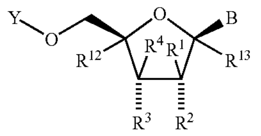
with the proviso that when R^a and R^c are α-hydroxy, R^e is amino, R^b is β-methyl and R^h is hydrogen or R^h is β-methyl and R^b is hydrogen, and R^f, R^g, R^i, R^k, and R^l are hydrogen, then R^d is not cyano or $CONH_2$.

The compounds of formula XII are also inhibitors of RNA-dependent RNA viral replication and in particular of HCV replication and are useful for the treatment of RNA-dependent RNA viral infection and in particular for the treatment of HCV infection.

Also encompassed within the present invention are pharmaceutical compositions containing the compounds alone or in combination with other agents active against RNA-dependent RNA virus and in particular against HCV.

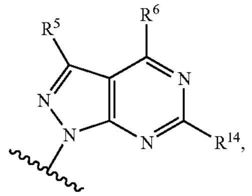
DETAILED DESCRIPTION OF THE INVENTION

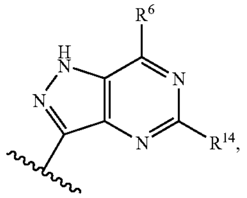
The present invention provides a method for inhibiting RNA-dependent RNA viral polymerase, a method for inhibiting RNA-dependent RNA viral replication, and/or a method for treating RNA-dependent RNA viral infection in a mammal in need thereof comprising administering to the mammal a therapeutically effective amount of a compound of structural formula I which is of the stereochemical configuration:

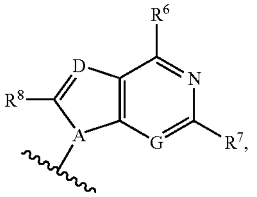


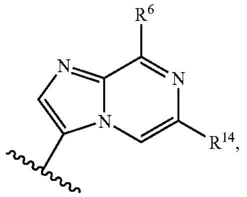
(I)

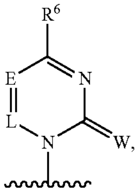
or a pharmaceutically acceptable salt thereof,
wherein B is selected from the group consisting of

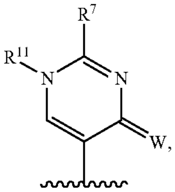




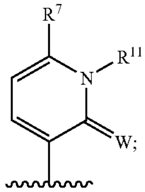








and



A, G, and L are each independently CH or N;

D is N, CH, C—CN, C—NO_2, C—C_{1-3} alkyl, C—$NHCONH_2$, C—$CONR^{11}R^{11}$, C—$CSNR^{11}R^{11}$, C—$COOR^{11}$, C—C(=NH)NH_2, C-hydroxy, C—C_{1-3} alkoxy, C-amino, C—C_{1-4} alkylamino, C-di(C_{1-4} alkyl)amino, C-halogen, C-(1,3-oxazol-2-yl), C-(1,3-thiazol-2-yl), or C-(imidazol-2-yl); wherein alkyl is unsubstituted or substituted with one to three groups independently selected from halogen, amino, hydroxy, carboxy, and C_{1-3} alkoxy;

E is N or CR^5;

W is O or S;

Y is H, C_{1-10} alkylcarbonyl, $P_3O_9H_4$, $P_2O_6H_3$, or P(O)R^9R^{10};

R^1 is hydrogen, C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms and one of R^2 and R^3 is hydroxy or C_{1-4} alkoxy and the other of R^2 and R^3 is selected from the group consisting of

hydrogen,
hydroxy,
halogen,
C_{1-4} alkyl, optionally substituted with 1 to 3 fluorine atoms,
C_{1-10} alkoxy, optionally substituted with C_{1-3} alkoxy or 1 to 3 fluorine atoms,
C_{2-6} alkenyloxy,
C_{1-4} alkylthio,
C_{1-8} alkylcarbonyloxy,
aryloxycarbonyl,
azido,
amino,
C_{1-4} alkylamino, and
di(C_{1-4} alkyl)amino; or

R^2 is hydrogen, C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms and one of R^1 and R^3 is hydroxy or C_{1-4} alkoxy and the other of R^1 and R^3 is selected from the group consisting of

hydrogen,
hydroxy,
halogen,
C_{1-4} alkyl, optionally substituted with 1 to 3 fluorine atoms;
C_{1-10} alkoxy, optionally substituted with hydroxy, C_{1-3} alkoxy, carboxy, or 1 to 3 fluorine atoms,
C_{2-6} alkenyloxy,
C_{1-4} alkylthio,
C_{1-8} alkylcarbonyloxy,
aryloxycarbonyl,
azido,
amino,
C_{1-4} alkylamino, and
di(C_{1-4} alkyl)amino; or

R^1 and R^2 together with the carbon atom to which they are attached form a 3- to 6-membered saturated monocyclic ring system optionally containing a heteroatom, selected from O, S, and NC_{0-4} alkyl;

R^4 and R^6 are each independently H, OH, SH, NH_2, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, C_{3-6} cycloalkylamino, halogen, C_{1-4}alkyl, C_{1-4} alkoxy, or CF_3;

R^5 is H, C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-4} alkylamino, CF_3, or halogen;

R^{14} is H, CF_3, C_{1-4} alkyl, amino, C_{1-4} alkylamino, C_{3-6} cycloalkylamino, or di(C_{1-4} alkyl)amino;

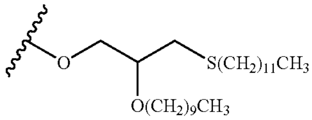
R^7 is hydrogen, amino, C_{1-4} alkylamino, C_{3-6} cycloalkylamino, or di(C_{1-4} alkyl)amino;

each R^{11} is independently H or C_{1-6} alkyl;

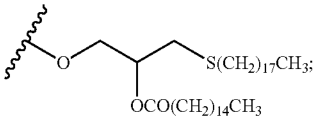
R^8 is H, halogen, CN, carboxy, C_{1-4} alkyloxycarbonyl, N_3, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, hydroxy, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} alkylsulfonyl, or (C_{1-4} alkyl)$_{0-2}$ aminomethyl;

R^{12} and R^{13} are each independently hydrogen, methyl, hydroxymethyl, or fluoromethyl; and

R^9 and R^{10} are each independently hydroxy, $OCH_2CH_2SC({=}O)C_{1-4}$ alkyl, $OCH_2O(C{=}O)OC_{1-4}$ alkyl, $NHCHMeCO_2Me$, $OCH(C_{1-4}$ alkyl$)O(C{=}O)C_{1-4}$ alkyl,

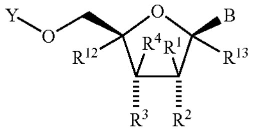


or



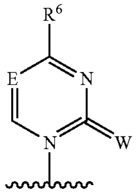
with the provisos that (a) when R^1 is hydrogen, one of R^3 and R^4 is hydrogen, and R^2 is fluoro, then the other of R3 and R^4 is not hydrogen, halogen, azido, trifluoromethyl, C_{1-4} alkyl, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, or C_{1-10} alkoxy; (b) when R^1 is hydrogen, one of R^3 and R^4 is hydrogen, and R^2 is halogen, hydroxy, C_{1-6} alkoxy, or C_{2-6} alkenyloxy, then the other of R^3 and R^4 is not hydrogen, fluoro, or azido; and (c) when R^1 and R^3 are hydrogen and R^2 is hydroxy, then R^4 is not hydroxy.

In one embodiment of the present invention is the method of inhibiting RNA-dependent RNA viral polymerase, inhibiting RNA-dependent viral replication, and/or treating RNA-dependent RNA viral infection with a compound of structural formula II which is of the stereochemical configuration:

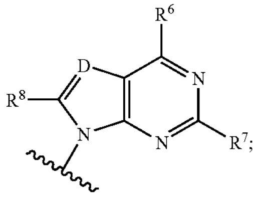


(II)

wherein B is



or



D is N, CH, C—CN, C—NO_2, C—C_{1-3} alkyl, C—$NHCONH_2$, C—$CONR^{11}R^{11}$, C—$CSNR^{11}R^{11}$, C—$COOR^{11}$, C-hydroxy, C—C_{1-3} alkoxy, C-amino, C—C_{1-4} alkylamino, C-di(C_{1-4} alkyl)amino, C-halogen, C-(1,3-oxazol-2-yl), C-(1,3-thiazol-2-yl), or C-(imidazol-2-yl);

wherein alkyl is unsubstituted or substituted with one to three groups independently selected from halogen, amino, hydroxy, carboxy, and C_{1-3} alkoxy;

E is N or C-R^5;

W is O or S;

Y is H, C_{1-10} alkylcarbonyl, $P_3O_9H_4$, or $P(O)R^9R^{10}$;

R^1 is hydrogen, CF_3, or C_{1-4} alkyl and one of R^2 and R^3 is OH or C_{1-4} alkoxy and the other of R^2 and R^3 is selected from the group consisting of

hydrogen,
hydroxy,
halogen,
C_{1-3} alkyl,
trifluoromethyl,
C_{1-4} alkoxy,
C_{1-4} alkylthio,
C_{1-8} alkylcarbonyloxy,
aryloxycarbonyl,
azido,
amino,
C_{1-4} alkylamino, and
di(C_{1-4} alkyl)amino; or

R^2 is hydrogen, CF_3, or C_{1-4} alkyl and one of R^1 and R^3 is OH or C_{1-4} alkoxy and the other of R^1 and R^3 is selected from the group consisting of

hydrogen,
hydroxy,
fluoro,
C_{1-4} alkyl,
trifluoromethyl,
C_{1-4} alkoxy,
C_{1-4} alkylthio,
C_{1-8} alkylcarbonyloxy,
azido,
amino,
C_{1-4} alkylamino, and
di(C_{1-4} alkyl)amino; or

R^1 and R^2 together with the carbon atom to which they are attached form a 3- to 6-membered saturated monocyclic ring system optionally containing a heteroatom selected from O, S, and NC_{0-4} alkyl;

R^4 and R^6 are each independently H, OH, SH, NH_2, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, C_{3-6} cycloalkylamino, halogen, C_{1-4} alkyl, C_{1-4} alkoxy, or CF_3;

R^5 is H, C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-4} alkylamino, CF_3, or halogen;

R^7 is hydrogen, amino, C_{1-4} alkylamino, C_{3-6} cycloalkylamino, or di(C_{1-4} alkyl)amino;

each R^{11} is independently H or C_{1-6} alkyl;

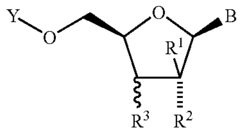
R^8 is H, halogen, CN, carboxy, C_{1-4} alkyloxycarbonyl, N_3, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, hydroxy, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} alkylsulfonyl, or (C_{1-4} alkyl)$_{0-2}$ aminomethyl;

R^{12} and R^{13} are each independently hydrogen or methyl; and

R^9 and R^{10} are each independently hydroxy, $OCH_2CH_2SC({=}O)C_{1-4}$ alkyl, or $OCH_2O(C{=}O)C_{1-4}$ alkyl;

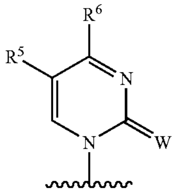
with the provisos that (a) when R^1 is hydrogen, one of R^3 and R^4 is hydrogen, and R^2 is fluoro, then the other of R^3 and R^4 is not hydrogen, halogen, trifluoromethyl, C_{1-4} alkyl, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, or C_{1-4} alkoxy; (b) when R^1 is hydrogen, one of R^3 and R^4 is hydrogen, and R^2 is halogen, hydroxy, or C_{1-4} alkoxy, then the other of R^3 and R^4 is not hydrogen, fluoro, or azido; and (c) when R^1 and R^3 are hydrogen and R^2 is hydroxy, then R^4 is not hydroxy.

In a second embodiment of the present invention is the method of inhibiting RNA-dependent RNA viral polymerase, inhibiting RNA-dependent RNA viral replication, and/or treating RNA-dependent RNA viral infection with a compound of structural formula III which is of the stereochemical configuration:

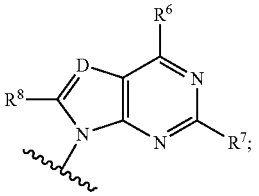


(III)

wherein B is



or



D is N, CH, C—CN, C—NO2, C—C_{1-3} alkyl, C—NHCONH$_2$, C—CONR^{11}R^{11}, C—CSNR^{11}R^{11}, C—COOR^{11}, C-hydroxy, C—C_{1-3} alkoxy, C-amino, C—C_{1-4} alkylamino, C-di(C_{1-4} alkyl)amino, C-halogen, C-(1,3-6oxazol-2-yl), C-(1,3-thiazol-2-yl), or C-(imidazol-2-yl); wherein alkyl is unsubstituted or substituted with one to three groups independently selected from halogen, amino, hydroxy, carboxy, and

C_{1-3} alkoxy;

W is O or S;

Y is H, C_{1-10} alkylcarbonyl, $P_3O_9H_4$, $P_2O_6H_3$, or P(O) R^{9}R^{10};

R^1 is hydrogen, CF$_3$, or C_{1-4} alkyl and one of R^2 and R^3 is OH or C_{1-4} alkoxy and the other of R^2 and R^3 is selected from the group consisting of

hydrogen,
hydroxy,
fluoro,
C_{1-3} alkyl,
trifluoromethyl,
C_{1-8} alkylcarbonyloxy,
C_{1-3} alkoxy, and
amino; or

R^2 is hydrogen, CF$_3$, or C_{1-4} alkyl and one of R^1 and R^3 is OH or C_{1-4} alkoxy and the other of R^1 and R^3 is selected from the group consisting of

hydrogen,
hydroxy,
fluoro,
C_{1-3} alkyl,
trifluoromethyl,
C_{1-8} alkylcarbonyloxy,
C_{1-3} alkoxy, and
amino; or

R^1 and R^2 together with the carbon atom to which they are attached form a 3- to 6-membered saturated monocyclic ring system optionally containing a heteroatom selected from O, S, and NC$_{0-4}$ alkyl;

R^6 is H, OH, SH, NH$_2$, C_{1-4} alkylamino, di(C_{1-4} alkyl) amino, C_{3-6} cycloalkylamino, halogen, C_{1-4} alkyl, C_{1-4} alkoxy, or CF$_3$;

R^5 is H, C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-4} alkylamino, CF$_3$, or halogen;

R^7 is hydrogen, amino, C_{1-4} alkylamino, C_{3-6} cycloalkylamino, or di(C_{1-4} alkyl)amino;

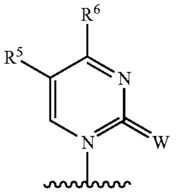
each R^{11} is independently H or C_{1-6} alkyl;

R^8 is H, halogen, CN, carboxy, C_{1-4} alkyloxycarbonyl, N$_3$, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, hydroxy, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} alkylsulfonyl, or (C_{1-4} alkyl)$_{0-2}$ aminomethyl; and

R^9 and R^{10} are each independently hydroxy, OCH$_2$CH$_2$SC (═O)t-butyl, or OCH$_2$O(C═O)iPr;

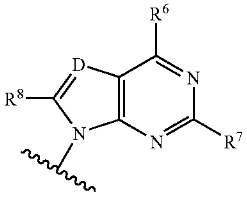
with the provisos that (a) when R^1 is hydrogen and R^2 is fluoro, then R^3 is not hydrogen, trifluoromethyl, fluoro, C_{1-3} alkyl, amino, or C_{1-3} alkoxy; (b) when R^1 is hydrogen and R^2 is fluoro, hydroxy, or C_{1-3} alkoxy, then R^3 is not hydrogen or fluoro; and (c) when R^1 is hydrogen and R^2 is hydroxy, then R^3 is not β-hydroxy.

In a class of this embodiment is the method of inhibiting RNA-dependent RNA viral polymerase, inhibiting RNA-dependent RNA viral replication, and/or treating RNA-dependent RNA viral infection with a compound of structural formula III wherein B is



and W, Y, and the R substituents are as defined under this second embodiment.

In a second class of this embodiment is the method of inhibiting RNA-dependent RNA viral polymerase, inhibiting RNA-dependent RNA viral replication, and/or treating RNA-dependent RNA viral infection with a compound of structural formula m wherein B is



and Y, D, and the R substituents are as defined under this second embodiment.

In a third embodiment of the present-invention, the RNA-dependent RNA viral polymerase is a positive-sense single-stranded RNA-dependent RNA viral polymerase. In a class of this embodiment, the positive-sense single-stranded RNA-dependent RNA viral polymerase is a Flaviviridae viral polymerase or a Picornaviridae viral polymerase. In a subclass of this class, the Picornaviridae viral polymerase is rhinovirus polymerase, poliovirus polymerase, or hepatitis A virus polymerase. In a second subclass of this class, the Flaviviridae viral polymerase is selected from the group consisting of hepatitis C virus polymerase, yellow fever virus polymerase, dengue virus polymerase, West Nile virus polymerase, Japanese encephalitis virus polymerase, Banzi virus polymerase, and bovine viral diarrhea virus (BVDV) polymerase. In a subclass of this subclass, the Flaviviridae viral polymerase is hepatitis C virus polymerase.

In a fourth embodiment of the present invention, the RNA-dependent RNA viral replication is a positive-sense single-stranded RNA-dependent RNA viral replication. In a class of this embodiment, the positive-sense single-stranded RNA-dependent RNA viral replication is Flaviviridae viral replication or Picornaviridae viral replication. In a subclass of this class, the Picornaviridae viral replication is rhinovirus replication, poliovirus replication, or hepatitis A virus replication. In a second subclass of this classy the Flaviviridae viral replication is selected from the group consisting of hepatitis C virus replication, yellow fever virus replication, dengue virus replication, West Nile virus replication, Japanese encephalitis virus replication, Banzi virus replication, and bovine viral diarrhea virus replication. In a subclass of this subclass, the Flaviviridae viral replication is hepatitis C virus replication.

In a fifth embodiment of the present invention, the RNA-dependent RNA viral infection is a positive-sense single-stranded RNA-dependent viral infection. In a class of this embodiment, the positive-sense single-stranded RNA-dependent RNA viral infection is Flaviviridae viral infection or Picornaviridae viral infection. In a subclass of this class, the Picornaviridae viral infection is rhinovirus infection, poliovirus infection, or hepatitis A virus infection. In a second subclass of this class, the Flaviviridae viral infection is selected from the group consisting of hepatitis C virus infection, yellow fever virus infection, dengue virus infection, West Nile virus infection, Japanese encephalitis virus infection, Banzi virus infection, and bovine viral diarrhea virus infection. In a subclass of this subclass, the Flaviviridae viral infection is hepatitis C virus infection.

Illustrative of the invention is a method for inhibiting RNA-dependent RNA viral polymerase, inhibiting RNA-dependent RNA viral replication, and/or treating RNA-dependent RNA viral infection wherein the compound is selected from:

2'-O-methyl-cytidine,
2'-C-methyl-cytidine,
3',5'-di-O-octanoyl-2'-O-methyl-cytidine,
3'-O-octanoyl-2'-O-methyl-cytidine,
2'-C-methyl-adenosine,
8-amino-2'-C-methyladenosine,
4-amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile,
4-amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carboxamide,
3'-deoxy-3'-methyl-cytidine,
4-amino-7-(3-deoxy-3-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide,
3'-deoxy-adenosine,
4-amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide,
3'-amino-3'-deoxyadenosine,
2-amino-3,4-dihydro-4-oxo-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide,
4-amino-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide,
2-amino-3,4-dihydro-4-oxo-7-(13-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile,
2-amino-5-ethyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
6-amino-1-(β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one, 3'-deoxyguanosine,
2-amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2'-O-methylguanosine,
2-amino-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-7-(2-O-methyl-β-D-ribofuranosyl)-5H-pyrrolo[3,2-d]pyrimidin-4(3H)-one,
7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
3'-deoxycytidine,
2-amino-5-methyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-3,4-dihydro-4-oxo-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo-[2,3-d]pyrimidin-5-carbonitrile,
2-amino-5-methyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
8-azidoguanosine,
8-aminoguanosine,
8-bromoadenosine,
8-aminoadenosine,
8-bromoguanosine,
3'-deoxy-3'-fluorocytidine,
3'-deoxy-3'-fluoroguanosine,
4-amino-7-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide,
2-amino-4-chloro-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile,
2-amino-4-chloro-5-ethyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-4-chloro-5-methyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo-[2,3-d]pyrimidine,
2-amino-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-thione,
2-amino-4-chloro-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-4-chloro-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-4-chloro-5-methyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-]pyrimidine,
1-(β-D-ribofuranosyl)-1H-pyrazolo[3,4-d]pyrimidin-4(3H)-one,
4-amino-1-(β-D-ribofuranosyl)-1H-pyrazolo[3,4-d]pyrimidine,
2-amino-6-chloro-9-(β-D-ribofuranosyl)-9H-purine,
2-amino-4-chloro-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile,
6-methyl-9-(β-D-ribofuranosyl)-9H-purine,
2-amino-7-(2-deoxy-2-fluoro-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-4-chloro-7-(2-deoxy-2-fluoro-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-7-(β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-7-(β-D-arabinofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile,
2-amino-5-methyl-7-(β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
9-(β-D-arabinofuranosyl)-9H-purin-6(1H)-one,
1-(β-D-arabinofuranosyl)-1H-cytosine,
2-amino-4-chloro-5-methyl-7-(β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
3'-deoxy-3'-(fluoromethyl)-guanosine,
2'-amino-2'-deoxycytidine,
4-amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile,
2'-O-methyladenosine,
4-amino-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
3'-amino-3'-deoxy-2'-O-methyl-adenosine,

3'-deoxy-3'-methyl-uridine,
6-amino-1-(3-deoxy-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one,
6-amino-1-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(3H)-one,
3'-deoxy-3'-fluorouridine,
3'-deoxy-3'-fluoroadenosine,
2-amino-7-(2-deoxy-β-D-ribofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo[2,3-d]-pyrimidin-5-carbonitrile,
3'-deoxy-5-methyl-uridine,
3'-deoxy-2'-O-(2-methoxyethyl)-3'-methyl-5-methyluridine,
2'-amino-2'-deoxy-uridine,
2-amino-9-(β-D-arabinofuranosyl)-9H-purin-6(1H)-one,
3'-deoxy-3'-methylguanosine,
2'-O-[4-(imidazolyl-1)butyl]guanosine,
2'-deoxy-2'-fluoroguanosine,
2'-deoxyguanosine,
2-amino-7-(2-deoxy-2-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-7-(3-deoxy-β-D-ribofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile,
2-amino-5-iodo-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-7-(3-deoxy-3-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile,
2-amino-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-7-(2-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-7-(3-deoxy-3-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-7-(3-deoxy3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
6-amino-1-(2-O-methyl-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one,
6-amino-1-(2-deoxy-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one,
6-amino-1-(3-deoxy-3-methyl-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one,
6-amino-1-(2-deoxy-2-fluoro-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one,
6-amino-1-(β-D-arabinofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one,
2'-O-[2-(N,N-diethylaminooxy)ethyl]-5-methyluridine,
5-ethynyl-2'-O-(2-methoxyethyl)-cytidine,
1-(2-C-methyl-β-D-arabinofuranosyl)uracil,
5-methyl-3'-deoxycytidine,
2-amino-2'-O-methyladenosine,
2'-deoxy-2'-fluoroadenosine,
3'-deoxy-3'-fluoroadenosine,
3'-deoxy-3'-methyladenosine,
2-amino-7-(2-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-7-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide,
4-amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide,
4-amino-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-7-(β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-1-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridine,
4-amino-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine (tubercidin),
4,6-diamino-7-(13-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-7-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo-[2,3-d]pyrimidin-5-carboxamide,
4-amino-1-(3-deoxy-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridine,
4-amino-1-(3-deoxy-3-methyl-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridine,
4-amino-1-(β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridine,
4-amino-1-(2-C-methyl-β-D-ribofuranosyl)-1H-pyrazolo[3,4-d]pyrimidine,
4-amino-7-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine; and
the corresponding 5'-triphosphates, 5'-[bis(isopropyloxycarbonyloxymethyl)]monophosphates, 5'-mono-(S-C_{1-4} alkanoyl-2-thioethyl)monophosphates, and 5'-bis-(S-C_{1-4} alkanoyl-2-thioethyl)monophosphates thereof;

or a pharmaceutically acceptable salt thereof.

Further illustrative of the invention is a method for inhibiting RNA-dependent RNA viral polymerase, inhibiting RNA-dependent RNA viral replication, and/or treating RNA-dependent RNA viral infection wherein the compound is selected from:

2'-O-methyl-cytidine,
2'-C-methyl-cytidine,
3',5'-di-O-octanoyl-2'-O-methyl-cytidine,
3'-O-octanoyl-2'-O-methyl-cytidine,
4-amino-1-(β-D-ribofuranosyl)-1H-pyrazolo[3,4-d]pyrimidine,
4-amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile,
4-amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carboxamide,
2'-C-methyladenosine,
8-amino-2'-C-methyladenosine,
3'-deoxy-3'-methyl-cytidine,
4-amino-7-(3-deoxy-3-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide,
3'-deoxyadenosine,
4-amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide,
3'-amino-3'-deoxyadenosine,
2-amino-3,4-dihydro-4-oxo-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide,
4-amino-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide,
2-amino-3,4-dihydro-4-oxo-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile,
2-amino-5-ethyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
6-amino-1-(β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one,
3'-deoxyguanosine,
2-amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2'-O-methylguanosine,
2-amino-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-7-(2-O-methyl-β-D-ribofuranosyl)-5H-pyrrolo[3,2-d]pyrimidin-4-(3H)-one,
7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
3'-deoxy-cytidine,
2-amino-5-methyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-3,4-dihydro-4-oxo-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo-[2,3-d]pyrimidine-5-carbonitrile,

2-amino-5-methyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
8-azidoguanosine,
8-aminoguanosine,
8-bromoadenosine,
8-aminoadenosine,
8-bromoguanosine,
3'-deoxy-3'-fluorocytidine,
3'-deoxy-3'-fluoroguanosine,
4-amino-7-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]-pyrimidin-5-carboxamide,
2-amino-4-chloro-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile,
2-amino-4-chloro-5-ethyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-4-chloro-5-methyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo-[2,3-d]pyrimidine,
2-amino-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-4-chloro-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-4-chloro-5-methyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-7-(2-deoxy-2-fluoro-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
4-amino-1-(2-C-methyl-β-D-ribofuranosyl)-1H-pyrazolo[3,4-d]pyrimidine,
2-amino-7-(β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one, and
2-amino-7-(β-D-arabinofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile; and
the corresponding 5'-triphosphates, 5'-[bis(isopropyloxycarbonyloxymethyl)]monophosphates, 5'-mono-(S-pivaloyl-2-thioethyl)monophosphates, and 5'-bis-(S-pivaloyl-2-thioethyl)monophosphates thereof;
or a pharmaceutically acceptable salt thereof.

Even further illustrative of the present invention is a method for inhibiting RNA-dependent RNA viral polymerase, inhibiting RNA-dependent RNA viral replication, and/or treating RNA-dependent RNA viral infection wherein the compound is selected from

2'-O-methyl-cytidine,
2'-C-methyl-cytidine,
3',5'-di-O-octanoyl-2'-O-methyl-cytidine,
3'-O-octanoyl-2'-O-methyl-cytidine,
4-amino-1-(β-D-ribofuranosyl)-1H-pyrazolo[3,4-d]pyrimidine,
4-amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile,
4-amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carboxamide,
2'-C-methyladenosine,
8-amino-2'-C-methyladenosine,
8-bromoguanosine,
8-aminoguanosine,
8-aminoadenosine,
4-amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-4-chloro-5-ethyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-3,4-dihydro-4-oxo-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide,
4-amino-1-(2-C-methyl-β-D-ribofuranosyl)-1H-pyrazolo[3,4-d]pyrimidine,
2-amino-4-chloro-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile;
and the corresponding 5'-triphosphates thereof;

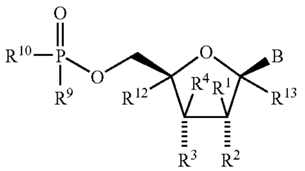
2'-O-methylcytidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate],
2-amino-7-(3-deoxy-β-D-ribofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo[2,3-d]pyrimidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate],
3'-deoxyguanosine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate], and
3'-deoxycytidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate];
or a pharmaceutically acceptable salt thereof.

Yet further illustrative of the invention is a method for inhibiting RNA-dependent RNA viral polymerase, inhibiting RNA-dependent RNA viral replication, and/or treating RNA-dependent RNA viral infection wherein the compound is selected from:

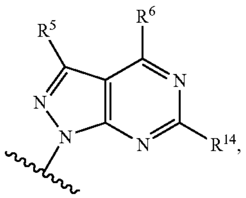
2'-O-methylcytidine,
2'-C-methylcytidine,
3',5'-di-O-octanoyl-2'-O-methyl-cytidine,
3'-O-octanoyl-2'-O-methyl-cytidine,
4-amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile,
4-amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carboxamide,
2'-C-methyladenosine,
8-amino-2'-C-methyladenosine,
2'-O-methylcytidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate],
2-amino-7-(3-deoxy-β-D-ribofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo[2,3-d]pyrimidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate], and
3'-deoxycytidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate];
or a pharmaceutically acceptable salt thereof.

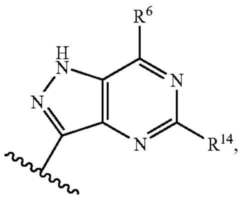
The present invention also provides novel compounds of structural formula IV of the indicated stereochemical configuration which are useful as inhibitors of RNA-dependent RNA viral polymerase:

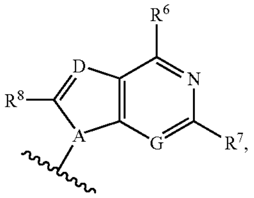
(IV)

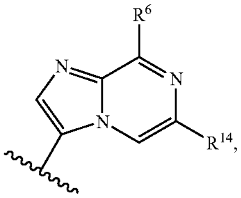


wherein B is selected from the group consisting of









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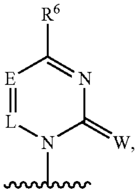
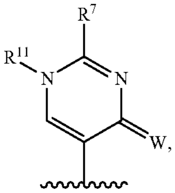
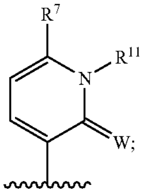




and



A, G, and L are each independently CH or N;

D is N, CH, C—CN, C—NO_2, C—C_{1-3} alkyl, C—$NHCONH_2$, C—$CONR^{11}R^{11}$, C—$CSNR^{11}R^{11}$, C—$COOR^{11}$, C-hydroxy, C—C_{1-3} alkoxy, C-amino, C—C_{1-4} alkylamino, C-di(C_{1-4} alkyl)amino, C-halogen, C-(1,3-oxazol-2-yl), C-(1,3-thiazol-2-yl), or C-(imidazol-2-yl); wherein alkyl is unsubstituted or substituted with one to three groups independently selected from halogen, amino, hydroxy, carboxy, and

C_{1-3} alkoxy;

E is N or CR^5;

W is O or S;

R^1 is hydrogen, C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms and one of R^2 and R^3 is hydroxy or C_{1-4} alkoxy and the other of R^2 and R^3 is selected from the group consisting of

- hydrogen,
- hydroxy,
- halogen,
- C_{1-4} alkyl, optionally substituted with 1 to 3 fluorine atoms,
- C_{1-10} alkoxy, optionally substituted with C_{1-3} alkoxy of 1 to 3 fluorine atoms,
- C_{2-6} alkenyloxy,
- C_{1-4} alkylthio,
- C_{1-8} alkylcarbonyloxy,
- aryloxycarbonyl,
- azido,
- amino,
- C_{1-4} alkylamino, and
- di(C_{1-4} alkyl)amino; or

R^2 is hydrogen, C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms and one of R^1 and R^3 is hydroxy or C_{1-4} alkoxy and the other of R^1 and R^3 is selected from the group consisting of

- hydrogen,
- hydroxy,
- halogen,
- C_{1-4} alkyl, optionally substituted with 1 to 3 fluorine atoms,
- C_{1-10} alkoxy, optionally substituted with hydroxy, C_{1-3} alkoxy, carboxy, or 1 to 3 fluorine atoms,
- C_{2-6} alkenyloxy,
- C_{1-4} alkylthio,
- C_{1-8} alkylcarbonyloxy,
- aryloxycarbonyl,
- azido,
- amino,
- C_{1-4} alkylamino, and
- di(C_{1-4} alkyl)amino; or

R^1 and R^2 together with the carbon atom to which they are attached form a 3- to 6-membered saturated monocyclic ring system optionally containing a heteroatom selected from O, S, and NC_{0-4} alkyl;

R^4 and R^6 are each independently H, OH, SH, NH_2, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, C_{3-6} cycloalkylamino, halogen, C_{1-4} alkyl, C_{1-4} alkoxy, or CF_3;

R^5 is H, C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-4} alkylamino, CF_3, or halogen;

R^{14} is H, CF_3, C_{1-4} alkyl, amino, C_{1-4} alkylamino, C_{3-6} cycloalkylamino, or di(C_{1-4} alkyl)amino;

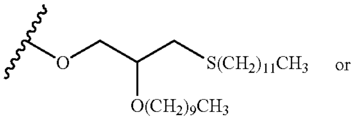
R^7 is hydrogen, amino, C_{1-4} alkylamino, C_{3-6} cycloalkylamino, or di(C_{1-4} alkyl)amino;

each R^{11} is independently H or C_{1-6} alkyl;

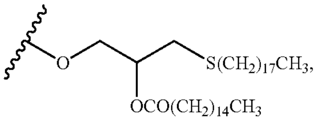
R^8 is H, halogen, CN, carboxy, C_{1-4} alkyloxycarbonyl, N_3, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, hydroxy, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} alkylsulfonyl, or (C_{1-4} alkyl)$_{0-2}$ aminomethyl;

R^{12} and R^{13} are each independently hydrogen or methyl; and

R^9 and R^{10} are each independently hydroxy, $OCH_2CH_2SC(=O)C_{1-4}$ alkyl, $OCH_2O(C=O)OC_{1-4}$ alkyl, $NHCHMeCO_2Me$, $OCH(C_{1-4}$ alkyl$)O(C=O)C_{1-4}$ alkyl,



or

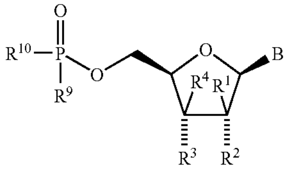


provided that at least one of R^9 and R^{10} is not hydroxy.

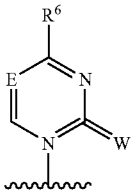
The compounds of formula IV are also inhibitors of RNA-dependent RNA viral replication and are useful for the treatment of RNA-dependent RNA viral infection.

In one embodiment, there are provided novel compounds of structural formula V which are of the stereochemical configuration:

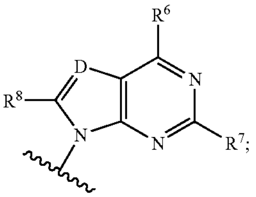
(V)



wherein B is



or



D is N, CH, C—CN, C—NO_2, C—C_{1-3} alkyl, C—$NHCONH_2$, C—$CONR^{11}R^{11}$, C—$CSNR^{11}R^{11}$, C—$COOR^{11}$, C-hydroxy, C—C_{1-3} alkoxy, C-amino, C—C_{1-4} alkylamino, C-di(C_{1-4} alkyl)amino, C-halogen, C-(1,3-oxazol-2-yl), C-(1,3-thiazol-2-yl), or C-(imidazol-2-yl); wherein alkyl is unsubstituted or substituted with one to three groups independently selected from halogen, amino, hydroxy, carboxy, and

C_{1-3} alkoxy;

W is O or S;

E is N or C-R^5;

R^1 is hydrogen, C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms and one of R^2 and R^3 is hydroxy or C_{1-4} alkoxy and the other of R^2 and R^3 is selected from the group consisting of
  hydrogen,
  hydroxy,
  halogen,
  C_{1-3} alkyl,
  trifluoromethyl,
  C_{1-4} alkoxy,
  C_{1-4} alkylthio,
  C_{1-8} alkylcarbonyloxy,
  aryloxycarbonyl,
  azido,
  amino,
  C_{1-4} alkylamino, and
  di(C_{1-4} alkyl)amino, or,

R^2 is hydrogen, C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms and one of R^1 and R^3 is hydroxy or C_{1-4} alkoxy and the other of R^1 and R^3 is selected from the group consisting of
  hydrogen,
  hydroxy,
  fluoro,
  C_{1-4} alkyl,
  trifluoromethyl,
  C_{1-4} alkoxy,
  C_{1-4} alkylthio,
  C_{1-8} alkylcarbonyloxy,
  azido,
  amino,
  C_{1-4} alkylamino, and
  di(C_{1-4} alkyl)amino; or

R^1 and R^2 together with the carbon atom to which they are attached form a 3- to 6-membered saturated monocyclic ring system optionally containing a heteroatom selected from O, S, and NC_{0-4} alkyl;

R^4 and R^6 are each independently H, OH, SH, NH_2, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, C_{3-6} cycloalkylamino, halogen, C_{1-4} alkyl, C_{1-4} alkoxy, or CF_3;

R^5 is H, C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-4} alkylamino, CF_3, or halogen;

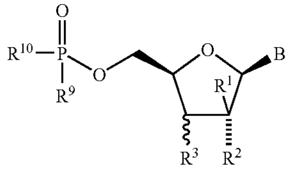
R^7 is hydrogen, amino, C_{1-4} alkylamino, C_{3-6} cycloalkylamino, or di(C_{1-4} alkyl)amino;

each R^{11} is independently H or C_{1-6} alkyl;

R^8 is H, halogen, CN, carboxy, C_{1-4} alkyloxycarbonyl, N_3, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, hydroxy, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} alkylsulfonyl, or
(C_{1-4} alkyl)$_{0-2}$ aminomethyl; and

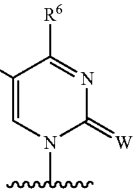
R^9 and R^{10} are each independently hydroxy, $OCH_2CH_2SC({=}O)C_{1-4}$ alkyl, or $OCH_2O(C{=}O)C_{1-4}$ alkyl, provided that at least one of R^9 and R^{10} is not hydroxy.

In a second embodiment, there are provided novel compounds of structural formula VI:

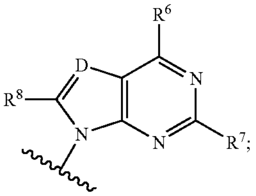


(VI)

wherein B is



or



D is N, CH, C—CN, C—NO_2, C—C_{1-3} alkyl, C—$NHCONH_2$, C—$CONR^{11}R^{11}$, C—$CSNR^{11}R^{11}$, C—$COOR^{11}$, C-hydroxy, C—C_{1-3} alkoxy, C-amino, C—C_{1-4} alkylamino, C-di(C_{1-4} alkyl)amino, C-halogen, C-(1,3-oxazol-2-yl), C-(1,3-thiazol-2-yl), or C-(imidazol-2-yl); wherein alkyl is unsubstituted or substituted with one to three groups independently selected from halogen, amino, hydroxy, carboxy, and

C_{1-3} alkoxy;

W is O or S;

R^1 is hydrogen, C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms and one of R^2 and R^3 is hydroxy or C_{1-4} alkoxy and the other of R^2 and R^3 is selected from the group consisting of
  hydrogen,
  hydroxy,
  fluoro,
  C_{1-3} alkyl,
  trifluoromethyl,
  C_{1-3} alkoxy,
  C_{1-8} alkylcarbonyloxy, and
  amino; or

R^2 is hydrogen, C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms and one of R^1 and R^3 is hydroxy or C_{1-4} alkoxy and the other of R^1 and R^3 is selected from the group consisting of
  hydrogen,
  hydroxy,
  fluoro,
  C_{1-3} alkyl,
  trifluoromethyl,
  C_{1-3} alkoxy,
  C_{1-8} alkylcarbonyloxy, and
  amino; or

R^1 and R^2 together with the carbon atom to which they are attached form a 3- to 6-membered saturated monocyclic ring system optionally containing a heteroatom selected from O, S, and NC_{0-4} alkyl,

R^6 is H, OH, SH, NH_2, C_{1-4} alkylamino, di(C_{1-4} alkyl) amino, C_{3-6} cycloalkylamino, halogen, C_{1-4} alkyl, C_{1-4} alkoxy, or CF_3;

R^5 is H, C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-4} alkylamino, CF_3, or halogen;

R^7 is hydrogen, amino, C_{1-4} alkylamino, C_{3-6} cycloalkylamino, or di(C_{1-4} alkyl)amino;

each R^{11} is independently H or C_{1-6} alkyl;

R^8 is H, halogen, CN, carboxy, C_{1-4} alkyloxycarbonyl, N_3, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, hydroxy, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} alkylsulfonyl, or (C_{1-4} alkyl)$_{0-2}$ aminomethyl; and

R^9 and R^{10} are each independently hydroxy, $OCH_2CH_2SC(=O)$t-butyl, or $OCH_2O(C=O)$iPr, provided that at least one of R^9 and R^{10} is not hydroxy.

Illustrative of the novel compounds of structural formula VI of the present invention are the following:

2'-O-methylcytidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate],

2-amino-7-(3-deoxy-β-D-ribofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo[2,3-d]pyrimidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate],

3'-deoxyguanosine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate],

2'-O-methylguanosine-5'-[bis-(S-acetyl-2-thioethyl)phosphate],

2'-O-methylguanosine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate],

8-bromo-2'-O-methylguanosine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate],

2-amino-3,4-dihydro-7-(2-O-methyl-β-D-ribofuranosyl)4-oxo-7H-pyrrolo[2,3-d]pyrimidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate],

2-amino-5-bromo-7-(3-deoxy-β-D-ribofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo[2,3-d]pyrimidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate],

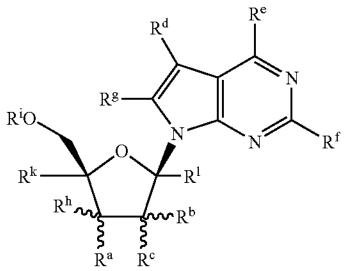
5-bromo-2'-O-methylcytidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate],

3-deoxycytidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate], and

2'-O-methylcytidine-5'-[bis(isopropyloxycarbonyloxymethyl)]phosphate.

The present invention further provides novel compounds of structural formula XII of the indicated stereochemical configuration or a pharmaceutically acceptable salt thereof which are useful as inhibitors of RNA-dependent RNA viral polymerase:

(XII)



wherein R^a and R^h are each independently selected from the group consisting of hydrogen, cyano, azido, halogen, hydroxy, mercapto, amino, C_{1-4} alkoxy, C_{2-4} alkenyl, C_{2-4} alkynyl, and C_{1-4} alkyl, wherein alkyl is unsubstituted or substituted with hydroxy, amino, C_{1-4} alkoxy, C_{1-4} alkylthio, or one to three fluorine atoms;

R^b is C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl, wherein alkyl is unsubstituted or substituted with hydroxy, amino, C_{1-4} alkoxy, C_{1-4} alkylthio, or one to three fluorine atoms;

R^c is hydrogen, fluorine, hydroxy, mercapto, C_{1-4} alkoxy, or C_{1-4} alkyl; or R^b and R^c together with the carbon atom to which they are attached form a 3- to 6-membered saturated monocyclic ring system optionally containing a heteroatom selected from O, S, and NC_{0-4} alkyl;

R^d is hydrogen, cyano, nitro, C_{1-3} alkyl, $NHCONH_2$, CONRjRj, CSNRjRj, COORj, $C(=NH)NH_2$, hydroxy, C_{1-3} alkoxy, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, halogen, (1,3-oxazol-2-yl), (1,3-thiazol-2-yl), or (imidazol-2-yl); wherein alkyl is unsubstituted or substituted with one to three groups independently selected from halogen, amino, hydroxy, carboxy, and C_{1-3} alkoxy;

R^e and R^f are each independently hydrogen, hydroxy, halogen, C_{1-4} alkoxy, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, C_{3-6} cycloalkylamino, di(C_{3-6} cycloalkyl)amino, or C_{4-6} cycloheteroalkyl, unsubstituted or substituted with one to two groups independently selected from halogen, hydroxy, amino, C_{1-4} alkyl, and C_{1-4} alkoxy;

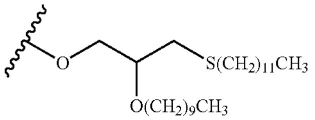
R^g is hydrogen, C_{1-4} alkyl, C_{2-4} alkynyl, halogen, cyano, carboxy, C_{1-4} alkyloxycarbonyl, azido, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, hydroxy, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} alkylsulfonyl, (C_{1-4} alkyl)$_{0-2}$ aminomethyl, or C_{4-6} cycloheteroalkyl, unsubstituted or substituted with one to two groups independently selected from halogen, hydroxy, amino, C_{1-4} alkyl, and C_{1-4} alkoxy;

R^i is hydrogen, C_{1-10} alkylcarbonyl, $P_3O_9H_4$, $P_2O_6H_3$, or $P(O)R^mR^n$;

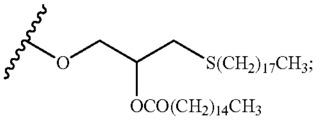
each R^j is independently hydrogen or C_{1-6} alkyl;

R^k and R^l are each independently hydrogen, methyl, hydroxymethyl, or fluoromethyl; and

R^m and R^n are each independently hydroxy, $OCH_2CH_2SC(=O)C_{1-4}$ alkyl, $OCH_2O(C=O)OC_{1-4}$ alkyl, $NHCHMeCO_2Me$, $OCH(C_{1-4}$ alkyl$)O(C=O)C_{1-4}$ alkyl,



or

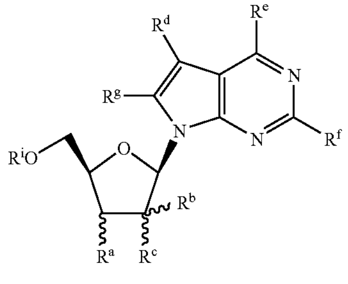


with the proviso that when R^a and R^c are α-hydroxy, R^e is amino, R^b is β-methyl and R^h is hydrogen or R^h is β-methyl and R^b is hydrogen, and R^f, R^g, R^i, R^k, and R^l are hydrogen, then R^d is not cyano or $CONH_2$.

The compounds of formula XII are also inhibitors of RNA-dependent RNA viral replication and are useful for the treatment of RNA-dependent RNA viral infection.

In one embodiment of the novel compounds of structural formula XII are the compounds of structural formula XIII:

XIII



wherein R^a is hydrogen, halogen, hydroxy, amino, or C_{1-3} alkoxy;

R^b is C_{1-3} alkyl, wherein alkyl is unsubstituted or substituted with hydroxy, amino, C_{1-3} alkoxy, C_{1-3} alkylthio, or one to three fluorine atoms;

R^c is hydroxy, fluoro, or C_{1-4} alkoxy;

R^d is hydrogen, cyano, methyl, halogen, or $CONH_2$;

R^g is hydrogen, amino, or C_{1-4} alkylamino;

R^i is hydrogen, $P_3O_9H_4$, $P_2O_6H_3$, or PO_3H_2; and

R^e and R^f are each independently hydrogen, hydroxy, halogen, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, or C_{3-6} cycloalkylamino;

with the proviso that when R^a and R^c are α-hydroxy, R^e is amino, R^b is methyl, and R^f, R^g, and R^i are hydrogen, then R^d is not cyano or $CONH_2$.

In a second embodiment of the compounds of structural formula XII are the compounds of structural formula XIII wherein:

R^b is methyl, fluoromethyl, hydroxymethyl, difluoromethyl, trifluoromethyl, or aminomethyl;

R^c is hydroxy, fluoro, or methoxy;

R^a is hydrogen, fluoro, hydroxy, amino, or methoxy;

R^i is hydrogen or $P_3O_9H_4$;

R^g is hydrogen or amino;

R^d is hydrogen, cyano, methyl, halogen, or $CONH_2$; and

R^e and R^f are each independently hydrogen, fluoro, hydroxy, or amino;

with the proviso that when R^b is β-methyl, R^a and R^c are α-hydroxy, R^e is amino, and

R^f, R^g, and R^i are hydrogen, then R^d is not cyano or $CONH_2$.

Illustrative of the novel compounds of the present invention of structural formula XIII which are useful as inhibitors of RNA-dependent RNA viral polymerase are the following:

4-amino-7-(2-C-methyl-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-methylamino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-dimethylamino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-cyclopropylamino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-7-(2-C-vinyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-7-(2-C-hydroxymethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-7-(2-C-fluoromethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-5-methyl-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carboxylic acid,
4-amino-5-bromo-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-5-chloro-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-5-fluoro-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2,4-diamino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-4-cyclopropylamino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
4-amino-7-(2-C-ethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-7-(2-C,2-O-dimethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-5-methyl-7-(2-C,2-O-dimethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
4-amino-7-(3-deoxy-2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-7-(3-deoxy-2-C-methyl-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-2-fluoro-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-7-(3-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-7-(3-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-7-(2,4-di-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine, and
4-amino-7-(3-deoxy-3-fluoro-2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine;

and the corresponding 5'-triphosphates;

or a pharmaceutically acceptable salt thereof.

Further illustrative of the novel compounds of the present invention of structural formula XIII which are useful as inhibitors of RNA-dependent RNA viral polymerase are the following:

4-amino-7-(2-C-methyl-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-7-(2-C-fluoromethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-5-methyl-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-5-bromo-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-5-chloro-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-5-fluoro-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine, and
4-amino-7-(2-C,2-O-dimethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,

and the corresponding 5'-triphosphates;

or a pharmaceutically acceptable salt thereof.

Further structurally novel nucleoside derivatives of the present invention which are useful as inhibitors of RNA-dependent RNA viral polymerase are the following:

3'-deoxy-3'-methyl-cytidine,
3',5-di-O-octanoyl-2'-O-methyl-cytidine,
3'-O-octanoyl-2'-O-methyl-cytidine,
4-amino-7-(3-deoxy-3-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide,
2-amino-5-ethyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
-2-amino-7-(2-O-methyl-β-D-ribofuranosyl)-5H-pyrrolo[3,2-d]pyrimidin-4(3H)-one,
7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-3,4-dihydro-4-oxo-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo-[2,3-d]pyrimidin-5-carbonitrile,
2-amino-5-methyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,

2-amino-4-chloro-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile,
2-amino-4-chloro-5-ethyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-4-chloro-5-methyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyridin-4(3H)-thione,
2-amino-4-chloro-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-4-chloro-5-methyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-7-(2-deoxy-2-fluoro-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-4-chloro-7-(2-deoxy-2-fluoro-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-7-(β-D-arabinofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile,
9-(β-D-arabinofuranosyl)-9H-purin-6(1H)-one,
3'-amino-3'-deoxy-2'-O-methyl-adenosine,
8-amino-2'-C-methyladenosine,
6-amino-1-(3-deoxy-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one,
6-amino-1-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(3H)-one,
3'-deoxy-2'-O-(2-methoxyethyl)-3'-methyl-5-methyluridine,
2-amino-7-(3-deoxy-β-D-ribofuranosyl)-3,4-dihydro-4oxo-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile,
2-amino-7-(3-deoxy-3-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyridin-5-carbonitrile,
2-amino-7-(3-deoxy-3-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyridin-4(3H)-one,
2-amino-7-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
6-amino-1-(2-O-methyl-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one,
6-amino-1-(3-deoxy-3-methyl-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one,
6-amino-1-(2-deoxy-2-fluoro-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one,
1-(2-C-methyl-β-D-arabinofuranosyl)uracil,
4-amino-1-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridine,
2-amino-7-(-3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo-[2,3-d]pyrimidin-5-carboxamide,
4-amino-1-(2-C-methyl-β-D-ribofuranosyl)-1H-pyrazolo[3,4-d]pyrimidine,
4-amino-1-(3-deoxy-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridine, and
4-amino-1-(3-deoxy-3-methyl-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridine;
and the corresponding 5'-triphosphates;
or a pharmaceutically acceptable salt thereof.

In a further embodiment the novel compounds of the present invention are useful as inhibitors of positive-sense single-stranded RNA-dependent RNA viral polymerase, inhibitors of positive-sense single-stranded RNA-dependent RNA viral replication, and/or for the treatment of positive-sense single-stranded RNA-dependent RNA viral infection. In a class of this embodiment, the positive-sense single-stranded RNA-dependent RNA virus is a Flaviviridae virus or a Picornaviridae virus. In a subclass of this class, the Picornaviridae virus is a rhinovirus, a poliovirus, or a hepatitis A virus. In a second subclass of this class, the Flaviviridae virus is selected from the group consisting of hepatitis C virus, yellow fever virus, dengue virus, West Nile virus, Japanese encephalitis virus, Banzi virus, and bovine viral diarrhea virus (BVDV). In a subclass of this subclass, the Flaviviridae virus is hepatitis C virus.

Throughout the instant application, the following terms have the indicated meanings:

The alkyl groups specified above are intended to include those alkyl groups of the designated length in either a straight or branched configuration. Exemplary of such alkyl groups are methyl, ethyl, propyl, isopropyl, butyl, sec-butyl, tertiary butyl, pentyl, isopentyl, hexyl, isohexyl, and the like.

The term “alkenyl” shall mean straight or branched chain alkenes of two to six total carbon atoms, or any number within this range (e.g., ethenyl, propenyl, butenyl, pentenyl, etc.).

The term “alkynyl” shall mean straight or branched chain alkynes of two to six total carbon atoms, or any number within this range (e.g., ethynyl, propynyl, butynyl, pentynyl, etc.).

The term “cycloalkyl” shall mean cyclic rings of alkanes of three to eight total carbon atoms, or any number within this range (i.e., cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, cycloheptyl, or cyclooctyl).

The term “cycloheteroalkyl” is intended to include non-aromatic heterocycles containing one or two heteroatoms selected from nitrogen, oxygen and sulfur. Examples of 4–6-membered cycloheteroalkyl include azetidinyl, pyrrolidinyl, piperidinyl, morpholinyl, thiamorpholinyl, imidazolidinyl, tetrahydrofuranyl, tetrahydropyranyl, tetrahydrothiophenyl, piperazinyl, and the like.

The term “alkoxy” refers to straight or branched chain alkoxides of the number of carbon atoms specified (e.g., C_{1-4} alkoxy), or any number within this range [i.e., methoxy (MeO—), ethoxy, isopropoxy, etc.].

The term “alkylthio” refers to straight or branched chain alkylsulfides of the number of carbon atoms specified (e.g., C_{1-4} alkylthio), or any number within this range [i.e., methylthio (MeS—), ethylthio, isopropylthio, etc.].

The term “alkylamino” refers to straight or branched alkylamines of the number of carbon atoms specified (e.g., C_{1-4} alkylamino), or any number within this range [i.e., methylamino, ethylamino, isopropylamino, t-butylamino, etc.].

The term “alkylsulfonyl” refers to straight or branched chain alkylsulfones of the number of carbon atoms specified (e.g., C_{1-6} alkylsulfonyl), or any number within this range [i.e., methylsulfonyl ($MeSO_2$—), ethylsulfonyl, isopropylsulfonyl, etc.].

The term “alkyloxycarbonyl” refers to straight or branched chain esters of a carboxylic acid derivative of the present invention of the number of carbon atoms specified (e.g., C_{1-4} alkyloxycarbonyl), or any number within this range [i.e., methyloxycarbonyl (MeOCO—), ethyloxycarbonyl, or butyloxycarbonyl].

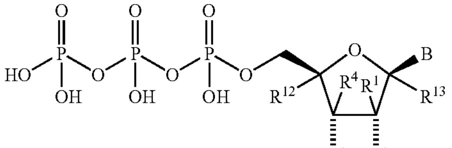
The term “aryl” includes both phenyl, naphthyl, and pyridyl. The aryl group is optionally substituted with one to three groups independently selected from C_{1-4} alkyl, halogen, cyano, nitro, trifluoromethyl, C_{1-4} alkoxy, and C_{1-4} alkylthio.

The term “halogen” is intended to include the halogen atoms fluorine, chlorine, bromine and iodine.

The term “substituted” shall be deemed to include multiple degrees of substitution by a named substituent. Where multiple substituent moieties are disclosed or claimed, the substituted compound can be independently substituted by one or more of the disclosed or claimed substituent moieties, singly or plurally.

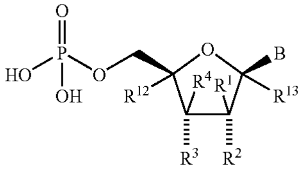
The term “5'-triphosphate” refers to a triphosphoric acid ester derivative of the 5'-hydroxyl group of a nucleoside compound of the present invention having the following general structural formula VII:

VII

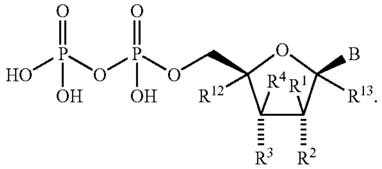


wherein B, Z, R^1-R^4, R^{12}, and R^{13} are as defined above. The compounds of the present invention are also intended to include pharmaceutically acceptable salts of the triphosphate ester as well as pharmaceutically acceptable salts of 5'-monophosphate and 5'-diphosphate ester derivatives of the structural formulae VIII and IX, respectively,

VIII

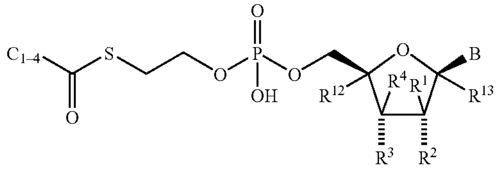


IX

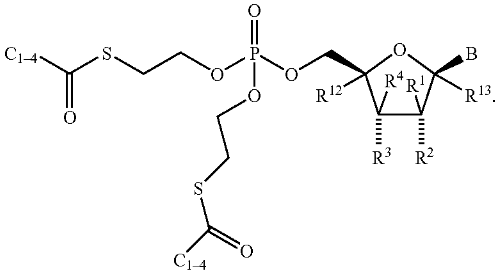


The term "5'-(S-acyl-2-thioethyl)phosphate" or "SATE" refers to a mono- or di-ester derivative of a 5'-monophosphate nucleoside of the present invention of structural formulae X and XI, respectively, as well as pharmaceutically acceptable salts of the mono-ester,

X



XI



The term "composition", as in "pharmaceutical composition," is intended to encompass a product comprising the active ingredient(s) and the inert ingredient(s) that make up the carrier, as well as any product which results, directly or indirectly, from combination, complexation or aggregation of any two or more of the ingredients, or from dissociation of one or more of the ingredients, or from other types of reactions or interactions of one or more of the ingredients. Accordingly, the pharmaceutical compositions of the present invention encompass any composition made by admixing a compound of the present invention and a pharmaceutically acceptable carrier.

The terms "administration of" and "administering a" compound should be understood to mean providing a compound of the invention or a prodrug of a compound of the invention to the individual in need.

Another aspect of the present invention is concerned with a method of inhibiting HCV NS5B polymerase, inhibiting HCV replication, or treating HCV infection with a compound of the present invention in combination with one or more agents useful for treating HCV infection. Such agents active against HCV include, but are not limited to, ribavirin, levovirin, viramidine, thymosin alpha-1, interferon-α, pegylated interferon-α peginterferon-α), a combination of interferon-α and ribavirin, a combination of peginterferon-α and ribavirin, a combination of interferon-α and levovirin, and a combination of peginterferon-α and levovirin. Interferon-α includes, but is not limited to, recombinant interferon-α2a (such as Roferon interferon available from Hoffman-LaRoche, Nutley, N.J.), pegylated interferon-α2a (Pegasys™), interferon-α2b (such as Intron-A interferon available from Schering Corp., Kenilworth, N.J.), pegylated interferon-α2b (PegIntron™), a recombinant consensus interferon (such as interferon alphacon-1), and a purified interferon-α product. Amgen's recombinant consensus interferon has the brand name Infergen®. Levovirin is the L-enantiomer of ribavirin which has shown immunomodulatory activity similar to ribavirin. Viramidine is an amino analog of ribavirin disclosed in WO 01/60379 (assigned to ICN Pharmaceuticals). In accordance with this method of the present invention, the individual components of the combination can be administered separately at different times during the course of therapy or concurrently in divided or single combination forms. The instant invention is therefore to be understood as embracing all such regimes of simultaneous or alternating treatment, and the term "administering" is to be interpreted accordingly. It will be understood that the scope of combinations of the compounds of this invention with other agents useful for treating HCV infection includes in principle any combination with any pharmaceutical composition for treating HCV infection. When a compound of the present invention or a pharmaceutically acceptable salt thereof is used in combination with a second therapeutic agent active against HCV, the dose of each compound may be either the same as or different from the dose when the compound is used alone.

For the treatment of HCV infection, the compounds of the present invention may also be administered in combination with an agent that is an inhibitor of HCV NS3 serine protease, such as LY570310 (VX-950). HCV NS3 serine protease is an essential viral enzyme and has been described to be an excellent target for inhibition of HCV replication. Both substrate and non-substrate based inhibitors of HCV NS3 protease inhibitors are disclosed in WO 98/17679, WO 98/22496, WO 98/46630, WO 99/07733, WO 99/07734, WO 99/38888, WO 99/50230, WO 99/64442, WO 00/09543, WO 00/59929, WO 01/74768, WO 01/81325, and GB-2337262. HCV NS3 protease as a target for the development of inhibitors of HCV replication and for the treatment of HCV infection is discussed in B. W. Dymock, "Emerging therapies for hepatitis C virus infection," *Emerging Drugs*, 6: 13–42 (2001).

Ribavirin, levovirin, and viramidine may exert their anti-HCV effects by modulating intracellular pools of guanine nucleotides via inhibition of the intracellular enzyme inosine monophosphate dehydrogenase (IMPDH). IMPDH is the rate-limiting enzyme on the biosynthetic route in de novo guanine nucleotide biosynthesis. Ribavirin is readily phosphorylated intracellularly and the monophosphate derivative is an inhibitor of IMPDH. Thus, inhibition of IMPDH represents another useful target for the discovery of inhibitors of HCV replication. Therefore, the compounds of the present invention may also be administered in combination with an inhibitor of IPDH, such as VX-497, which is disclosed in WO 97/41211 and WO 01/00622, (assigned to Vertex); another IMPDH inhibitor, such as that disclosed in WO 00/25780 (assigned to Bristol-Myers Squibb); or mycophenolate mofetil [see A. C. Allison and E. M. Eugui, *Agents Action*, 44 (Suppl.): 165 (1993)].

For the treatment of HCV infection, the compounds of the present invention may also be administered in combination with the antiviral agent amantadine (1-aminoadamaitane) [for a comprehensive description of this agent, see J. Kirschbaum, *Anal. Profiles Drug Subs*. 12: 1–36 (1983)].

The compounds of the present invention may also be combined for the treatment of HCV infection with antiviral 2'-C-branched ribonucleosides disclosed in R. E. Harry-O'kuru, et al., *J. Org. Chem*., 62: 1754–1759 (1997); M. S. Wolfe, et al., *Tetrahedron Lett*., 36: 7611–7614 (1995); and U.S. Pat. No. 3,480,613 (Nov. 25, 1969), the contents of which are incorporated by reference in their entirety. Such 2'-C-branched ribonucleosides include, but are not limited to, 2'-C-methyl-cytidine, 2'-C-methyl-adenosine, 2'-C-methyl-guanosine, and 9-(2-C-methyl-β-D-ribofuranosyl)-2,6-diaminopurine.

By "pharmaceutically acceptable" is meant that the carrier, diluent, or excipient must be compatible with the other ingredients of the formulation and not deleterious to the recipient thereof.

Also included within the present invention are pharmaceutical compositions comprising the novel nucleoside compounds and derivatives thereof of the present invention in association with a pharmaceutically acceptable carrier. Another example of the invention is a pharmaceutical composition made by combining any of the compounds described above and a pharmaceutically acceptable carrier. Another illustration of the invention is a process for making a pharmaceutical composition comprising combining any of the compounds described above and a pharmaceutically acceptable carrier.

Also included within the present invention are pharmaceutical compositions useful for inhibiting RNA-dependent RNA viral polymerase in particular HCV NS5B polymerase comprising an effective amount of a compound of this invention and a pharmaceutically acceptable carrier. Pharmaceutical compositions useful for treating RNA-dependent RNA viral infection in particular HCV infection are also encompassed by the present invention as well as a method of inhibiting RNA-dependent RNA viral polymerase in particular HCV NS5B polymerase and a method of treating RNA-dependent viral replication and in particular HCV replication. Additionally, the present invention is directed to a pharmaceutical composition comprising a therapeutically effective amount of a compound of the present invention in combination with a therapeutically effective amount of another agent active against RNA-dependent RNA virus and in particular against HCV. Agents active against HCV include, but are not limited to, ribavirin, levovirin, viramidine, thymosin alpha-1, an inhibitor of HCV NS3 serine protease, interferon-α, pegylated interferon-α peginterferon-α), a combination of interferon-α and ribavirin, a combination of peginterferon-α and ribavirin, a combination of interferon-α and levovirin, and a combination of peginterferon-α and levovirin. Interferon-α includes, but is not limited to, recombinant interferon-α2a (such as Roferon interferon available from Hoffmann-LaRoche, Nutley, N.J.), interferon-α2b (such as Intron-A interferon available from Schering Corp., Kenilworth, N.J.), a consensus interferon, and a purified interferon-α product. For a discussion of ribavirin and its activity against HCV, see J. O. Saunders and S. A. Raybuck, "Inosine Monophosphate Dehydrogenase: Consideration of Structure, Kinetics, and Therapeutic Potential," *Ann. Rep. Med. Chem*., 35: 201–210 (2000).

Another aspect of the present invention provides for the use of nucleoside compounds and derivatives thereof and their pharmaceutical compositions for the manufacture of a medicament for the inhibition of RNA-dependent RNA viral replication, in particular HCV replication, and/or the treatment of RNA-dependent RNA viral infection, in particular HCV infection. Yet a further aspect of the present invention provides for nucleoside compounds and derivatives thereof and their pharmaceutical compositions for use as a medicament for the inhibition of RNA-dependent RNA viral replication, in particular HCV replication, and/or for the treatment of RNA-dependent RNA viral infection, in particular HCV infection.

The pharmaceutical compositions of the present invention comprise a compound of structural formula I, IV, or XII as an active ingredient or a pharmaceutically acceptable salt thereof, and may also contain a pharmaceutically acceptable carrier and optionally other therapeutic ingredients.

The compositions include compositions suitable for oral, rectal, topical, parenteral (including subcutaneous, intramuscular, and intravenous), ocular (ophthalmic), pulmonary (nasal or buccal inhalation), or nasal administration, although the most suitable route in any given case will depend on the nature and severity of the conditions being treated and on the nature of the active ingredient. They may be conveniently presented in unit dosage form and prepared by any of the methods well-known in the art of pharmacy.

In practical use, the compounds of structural formulae I, IV, and XII can be combined as the active ingredient in intimate admixture with a pharmaceutical carrier according to conventional pharmaceutical compounding techniques. The carrier may take a wide variety of forms depending on the form of preparation desired for administration, e.g., oral, or parenteral (including intravenous). In preparing the compositions for oral dosage form, any of the usual pharmaceutical media may be employed, such as, for example, water, glycols, oils, alcohols, flavoring agents, preservatives, coloring agents and the like in the case of oral liquid preparations, such as, for example, suspensions, elixirs and solutions; or carriers such as starches, sugars, microcrystalline cellulose, diluents, granulating agents, lubricants, binders, disintegrating agents and the like in the case of oral solid preparations such as, for example, powders, hard and soft capsules and tablets, with the solid oral preparations being preferred over the liquid preparations.

Because of their ease of administration, tablets and capsules represent the most advantageous oral dosage unit form in which case solid pharmaceutical carriers are obviously employed. If desired, tablets may be coated by standard aqueous or nonaqueous techniques. Such compositions and preparations should contain at least 0.1 percent of active compound. The percentage of active compound in these compositions may, of course, be varied and may conveniently be between about 2 percent to about 60 percent of the weight of the unit. The amount of active compound in such therapeutically useful compositions is such that an effective dosage will be obtained. The active compounds can also be administered intranasally as, for example, liquid drops or spray.

The tablets, pills, capsules, and the like may also contain a binder such as gum tragacanth, acacia, corn starch or gelatin; excipients such as dicalcium phosphate; a disintegrating agent such as corn starch, potato starch, alginic acid; a lubricant such as magnesium stearate; and a sweetening agent such as sucrose, lactose or saccharin. When a dosage unit form is a capsule, it may contain, in addition to materials of the above type, a liquid carrier such as a fatty oil.

Various other materials may be present as coatings or to modify the physical form of the dosage unit. For instance, tablets may be coated with shellac, sugar or both. A syrup or elixir may contain, in addition to the active ingredient, sucrose as a sweetening agent, methyl and propylparabens as preservatives, a dye and a flavoring such as cherry or orange flavor.

Compounds of structural formulae I, IV, and XII may also be administered parenterally. Solutions or suspensions of these active compounds can be prepared in water suitably mixed with a surfactant such as hydroxy-propylcellulose. Dispersions can also be prepared in glycerol, liquid polyethylene glycols and mixtures thereof in oils. Under ordinary conditions of storage and use, these preparations contain a preservative to prevent the growth of microorganisms.

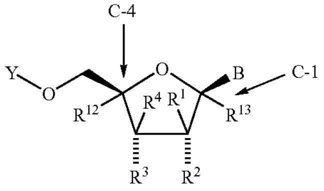
The pharmaceutical forms suitable for injectable use include sterile aqueous solutions or dispersions and sterile powders for the extemporaneous preparation of sterile injectable solutions or dispersions. In all cases, the form must be sterile and must be fluid to the extent that easy syringability exists. It must be stable under the conditions of manufacture and storage and must be preserved against the contaminating action of microorganisms such as bacteria and fungi. The carrier can be a solvent or dispersion medium containing, for example, water, ethanol, polyol (e.g. glycerol, propylene glycol and liquid polyethylene glycol), suitable mixtures thereof, and vegetable oils.

Any suitable route of administration may be employed for providing a mammal, especially a human with an effective dosage of a compound of the present invention. For example, oral, rectal, topical, parenteral, ocular, pulmonary, nasal, and the like may be employed. Dosage forms include tablets, troches, dispersions, suspensions, solutions, capsules, creams, ointments, aerosols, and the like. Preferably compounds of structural formulae I, IV, and XII are administered orally.

For oral administration to humans, the dosage range is 0.01 to 1000 mg/kg body weight in divided doses. In one embodiment the dosage range is 0.1 to 100 mg/kg body weight in divided doses. In another embodiment the dosage range is 0.5 to 20 mg/kg body weight in divided doses. For oral administration, the compositions are preferably provided in the form of tablets or capsules containing 1.0 to 1000 milligrams of the active ingredient, particularly, 1, 5, 10, 15, 20, 25, 50, 75, 100, 150, 200, 250, 300, 400, 500, 600, 750, 800, 900, and 1000 milligrams of the active ingredient for the symptomatic adjustment of the dosage to the patient to be treated.

The effective dosage of active ingredient employed may vary depending on the particular compound employed, the mode of administration, the condition being treated and the severity of the condition being treated. Such dosage may be ascertained readily by a person skilled in the art. This dosage regimen may be adjusted to provide the optimal therapeutic response.

The compounds of the present invention contain one or more asymmetric centers and can thus occur as racemates and racemic mixtures, single enantiomers, diastereomeric mixtures and individual diastereomers. The present invention is meant to comprehend nucleoside derivatives having the β-D stereochemical configuration for the five-membered furanose ring as depicted in the structural formula below, that is, nucleoside compounds in which the substituents at C-1 and C-4 of the five-membered furanose ring have the β-stereochemical configuration ("up" orientation as denoted by a bold line).

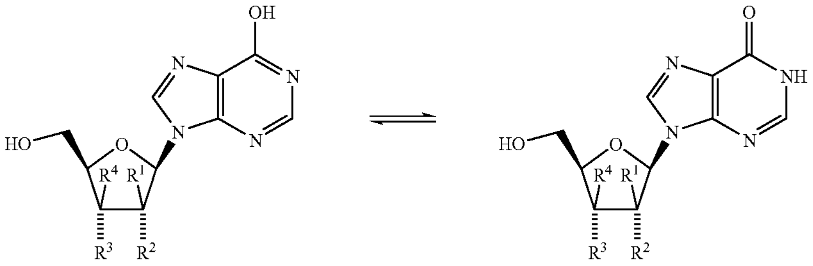


β-D-

The stereochemistry of the substituents at the C-2 and C-3 positions of the furanose ring of the compounds of the present invention is denoted either by a dashed line which signifies that the substituent, for example R^2 in structural formula VI, has the a (substituent "down") configuration or a squiggly line which signifies that the substituent, for example R^3 in structural formula VI, can have either the a (substituent "down") or β (substituent "up") configuration.

Some of the compounds described herein contain olefinic double bonds, and unless specified otherwise, are meant to include both E and Z geometric isomers.

Some of the compounds described herein may exist as tautomers such as keto-enol tautomers. The individual tautomers as well as mixtures thereof are encompassed with compounds of structural formulae I, IV, and XII. An example of keto-enol tautomers which are intended to be encompassed within the compounds of the present invention is illustrated below:

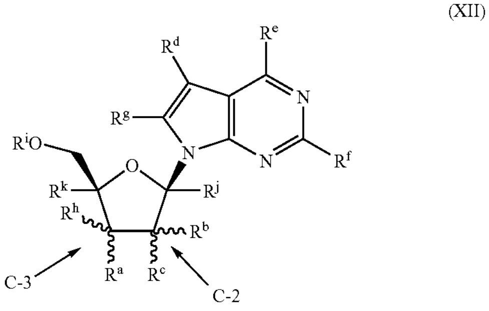


Compounds of structural formulae I, IV, and XII may be separated into their individual diastereoisomers by, for example, fractional crystallization from a suitable solvent, for example methanol or ethyl acetate or a mixture thereof, or via chiral chromatography using an optically active stationary phase.

Alternatively, any stereoisomer of a compound of the structural formulae I, IV, and XII may be obtained by stereospecific synthesis using optically pure starting materials or reagents of known configuration.

The stereochemistry of the substituents at the C-2 and C-3 positions of the furanose ring of the novel compounds of the present invention of structural formula XII is denoted by squiggly lines which signifies that substituents R^a, R^b, R^c and R^h can have either the α (substituent "down") or β (substituent "up") configuration independently of one another.

(XII)



The compounds of the present invention may be administered in the form of a pharmaceutically acceptable salt. The term "pharmaceutically acceptable salt" refers to salts prepared from pharmaceutically acceptable non-toxic bases or acids including inorganic or organic bases and inorganic or organic acids. Salts of basic compounds encompassed within the term "pharmaceutically acceptable salt" refer to non-toxic salts of the compounds of this invention which are generally prepared by reacting the free base with a suitable organic or inorganic acid. Representative salts of basic compounds of the present invention include, but are not limited to, the following: acetate, benzenesulfonate, benzoate, bicarbonate, bisulfate, bitartrate, borate, bromide, camsylate, carbonate, chloride, clavulanate, citrate, dihydrochloride, edetate, edisylate, estolate, esylate, fumarate, gluceptate, gluconate, glutamate, glycollylarsanilate, hexylresorcinate, hydrabamine, hydrobromide, hydrochloride, hydroxynaphthoate, iodide, isothionate, lactate, lactobionate, laurate, malate, maleate, mandelate, mesylate, methylbromide, methylnitrate, methylsulfate, mucate, napsylate, nitrate, N-methylglucamine ammonium salt, oleate, oxalate, pamoate (embonate), palmitate, pantothenate, phosphate/diphosphate, polygalacturonate, salicylate, stearate, sulfate, subacetate, succinate, tannate, tartrate, teoclate, tosylate, triethiodide and valerate. Furthermore, where the compounds of the invention carry an acidic moiety, suitable pharmaceutically acceptable salts thereof include, but are not limited to, salts derived from inorganic bases including aluminum, ammonium, calcium, copper, ferric, ferrous, lithium, magnesium, manganic, mangamous, potassium, sodium, zinc, and the like. Particularly preferred are the ammonium, calcium, magnesium, potassium, and sodium salts. Salts derived from pharmaceutically acceptable organic non-toxic bases include salts of primary, secondary, and tertiary amines, cyclic amines, and basic ion-exchange resins, such as arginine, betaine, caffeine, choline, N,N-dibenzylethylenediamine, diethylamine, 2-diethylaminoethanol, 2-dimethylaminoethanol, ethanolamine, ethylenediamine, N-ethylmorpholine, N-ethylpiperidine, glucamine, glucosamine, histidine, hydrabamine, isopropylamine, lysine, methylglucamine, morpholine, piperazine, piperidine, polyamine resins, procaine, purines, theobromine, triethylamine, trimethylamine, tripropylamine, tromethamine, and the like.

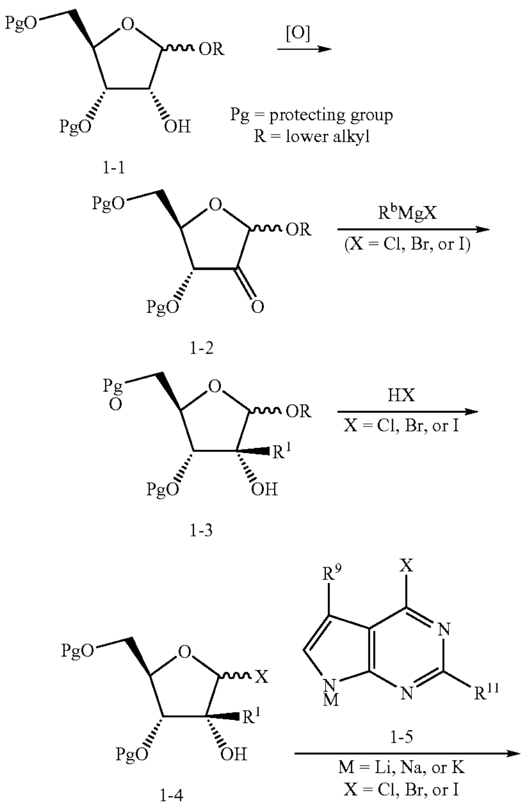
Also, in the case of a carboxylic acid (—COOH) or alcohol group being present in the compounds of the present invention, pharmaceutically acceptable esters of carboxylic acid derivatives, such as methyl, ethyl, or pivaloyloxymethyl, or acyl derivatives of alcohols, such as acetate or maleate, can be employed. Included are those esters and acyl groups known in the art for modifying the solubility or hydrolysis characteristics for use as sustained-release or prodrug formulations.

Preparation of the Nucleoside Compounds and Derivatives of the Invention

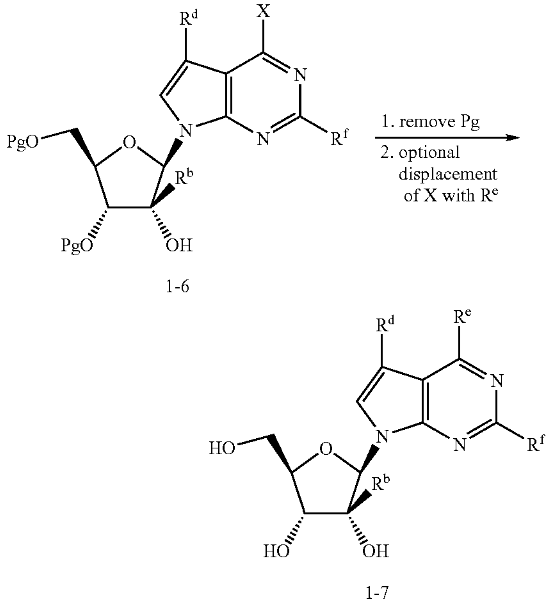
The nucleoside compounds and derivatives thereof of the present invention can be prepared following synthetic methodologies well-established in the practice of nucleoside and nucleotide chemistry. Reference is made to the following text for a description of synthetic methods used in the preparation of the compounds of the present invention: "Chemistry of Nucleosides and Nucleotides," L. B. Townsend, ed., Vols. 1–3, Plenum Press, 1988, which is incorporated by reference herein in its entirety.

A representative general method for the preparation of compounds of the present invention is outlined in Scheme 1 below. This scheme illustrates the synthesis of compounds of the present invention of structural formula 1-7 wherein the furanose ring has the β-D-ribo configuration. The starting material is a 3,5-bis-O-protected alkyl furanoside, such as methyl furanoside, of structural formula 1-1. The C-2 hydroxyl group is then oxidized with a suitable oxidizing agent, such as a chromium trioxide or chromate reagent or Dess-Martin periodinane, or by Swern oxidation, to afford a C-2 ketone of structural formula 1-2. Addition of a Grignard reagent, such as an alkyl, alkenyl, or alkynyl magnesium halide (for example, MeMgBr, EtMgBr, vinylMgBr, allylMgBr, and ethynylMgBr) or an alkyl, alkenyl, or alkynyl lithium, such as MeLi, across the carbonyl double bond of 1-2 in a suitable organic solvent, such as tetrahydrofuran, diethyl ether, and the like, affords the C-2 tertiary alcohol of structural formula 1-3. A good leaving group (such as Cl, Or, and I) is next introduced at the C-1 (anomeric) position of the furanose sugar derivative by treatment of the furanoside of formula 1-3 with a hydrogen halide in a suitable organic solvent, such as hydrogen bromide in acetic acid, to afford the intermediate furanosyl halide 1-4. A C-1 sulfonate, such methanesulfonate ($MeSO_2O$—, trifluoromethanesulfonate (CF_3SO_2O—), or p-toluenesulfonate (—OTs), may also serve as a useful leaving group in the subsequent reaction to generate the glycosidic (nucleosidic) linkage. The nucleosidic linkage is constructed by treatment of the intermediate of structural formula 1-4 with the metal salt (such as lithium, sodium, or potassium) of an appropriately substituted 1H-pyrrolo[2,3-d]pyrimidine 1-5, such as an appropriately substituted 4-halo-1H-pyrrolo[2,3-d]pyrimidine, which can be generated in situ by treatment with an alkali hydride (such as sodium hydride), an alkali hydroxide (such as potassium hydroxide), an alkali carbonate (such as potassium carbonate), or an alkali hexamethyldisilazide (such as NaHMDS) in a suitable anhydrous organic solvent, such as acetonitrile, tetrahydrofuran, 1-methyl-2-pyrrolidinone, or N,N-dimethylformamide (DMF). The displacement reaction can be catalyzed by using a phase-transfer catalyst, such as TDA-1 or triethylbenzylammonium chloride, in a two-phase system (solid-liquid or liquid-liquid). The optional protecting groups in the protected nucleoside of structural formula 1-6 are then cleaved following established deprotection methodologies, such as those described in T. W. Greene and P. G. M. Wuts, "Protective Groups in Organic Synthesis," 3^{rd} ed., John Wiley & Sons, 1999. Optional introduction of an amino group at the 4-position of the pyrrolo[2,3-d] pyrimidine nucleus is effected by treatment of the 4-halo intermediate 1-6 with the appropriate amine, such as alcoholic ammonia or liquid ammonia, to generate a primary amine at the C-4 position (—NH_2), an alkylamine to generate a secondary amine (—NHR), or a dialkylamine to generate a tertiary amine (—NRR'). A 7H-pyrrolo[2,3-d] pyrimidin-4(3H)one compound may be derived by hydrolysis of 1-6 with aqueous base, such as aqueous sodium hydroxide. Alcoholysis (such as methanolysis) of 1-6 affords a C-4 alkoxide (—OR), whereas treatment with an alkyl mercaptide affords a C-4 alkylthio (—SR) derivative. Subsequent chemical manipulations well-known to practitioners of ordinary skill in the art of organic/medicinal chemistry may be required to attain the desired compounds of the present invention.

Scheme 1



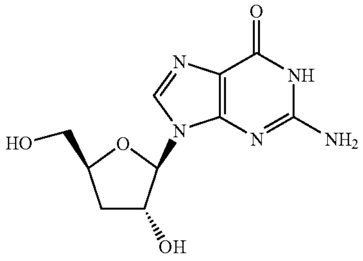
-continued



The examples below provide citations to literature publications, which contain details for the preparation of final compounds or intermediates employed in the preparation of final compounds of the present invention. The nucleoside compounds of the present invention were prepared according to procedures detailed in the following examples. The examples are not intended to be limitations on the scope of the instant invention in any way, and they should not be so construed. Those skilled in the art of nucleoside and nucleotide synthesis will readily appreciate that known variations of the conditions and processes of the following preparative procedures can be used to prepare these and other compounds of the present invention. All temperatures are degrees Celsius unless otherwise noted.

EXAMPLE 1

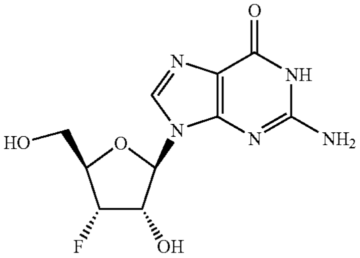
3'-Deoxyguanosine



This compound was prepared following the procedures described in *Nucleosides Nucleotides*, 13: 1049 (1994).

EXAMPLE 2

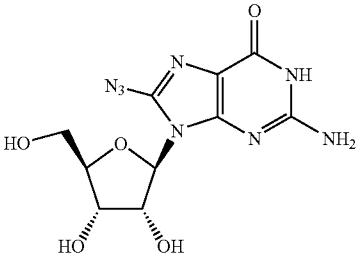
3'-Deoxy-3'-fluoroguanosine



This compound was prepared following the procedures described in *J. Med. Chem*. 34: 2195 (1991).

EXAMPLE 3

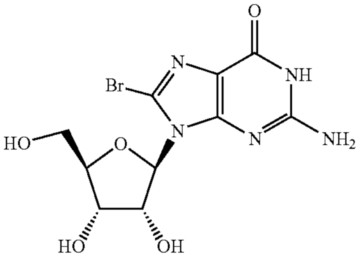
8-Azidoguanosine



This compound was prepared following the procedures described in *Chem. Pharm. Bull*..16: 1616 (1968).

EXAMPLE 4

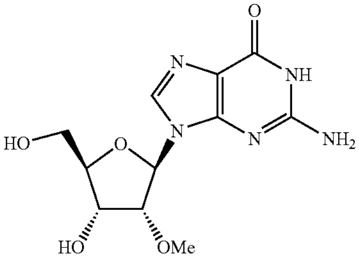
8-Bromoguanosine



This compound was obtained from commercial sources.

EXAMPLE 5

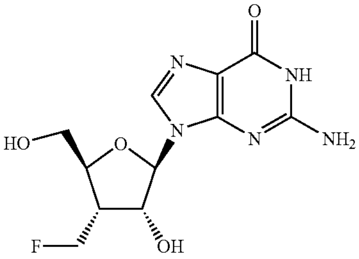
2'-O-Methylguanosine



This compound was obtained from commercial sources.

EXAMPLE 6

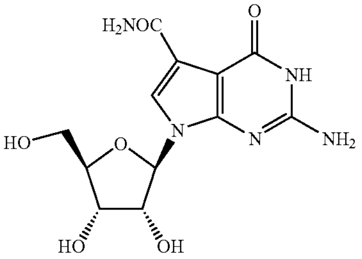
3'-Deoxy-3'-(fluoromethyl)guanosine



To a solution of 1,2-O-diacetyl-5-O-(p-toluoyl)-3-deoxy-3-(fluoromethyl)-D-ribofuranose (257 mg, 0.7 mmol) [prepared by a similar method as that described for the corresponding 5-O-benzyl derivative in *J Med. Chem*. 36: 353 (1993)] and N^2-acetyl-O^6-(diphenylcarbamoyl)guanine (554 mg, 1.43 mmol) in anhydrous acetonitrile (6.3 mL) was added bis(trimethylsilyl)acetamide (BSA) (1.03 g, 5 mmol). The reaction mixture was stirred at reflux for 30 minutes, and the bath was removed. The reaction mixture was cooled in an ice bath and TMS-triflate (288 mg, 1.3 mmol) was added with stirring. After addition was complete, the reaction was heated at reflux for 2 hr., the reaction mixture was poured onto ice and extracted with chloroform (5×10 mL). The combined organic layers were washed with aqueous saturated sodium bicarbonate, brine and dried over anhydrous Na_2SO_4. The solvent was removed under reduced pressure and the residue chromatographed over silica gel using 5% acetone/CH_2Cl_2 as the eluant to furnish the fully protected corresponding nucleoside derivative. This was dissolved in 1,4-dioxane (1.5 mL) to which was added 40% $MeNH_2/H_2O$ (1.3 g, 17 mmol). The reaction mixture was stirred for 1 day, evaporated and the residue crystallized with ether/MeOH to provide the title compound (58 mg). ^1H NMR (DMSO-d_6): δ 2.76–2.67 (m, 1H); 3.55–3.50 (m, 1H), 2.76–2.67 (m, 1H); 3.71–3.66 (m, 1H), 4.08–4.04 (m, 1H), 4.77–4.50 (m, 3H) 5.06 (t, 1H, J=5.3 Hz), 5.69 (d, 1H, J=3.4 Hz), 5.86 (d, 1H, J=5.1 Hz), 6.45 (bs, 2H), 7.97 (s, 1H), 10.59 (s, 1H). ^{19}F NMR (DMSO-d_6): δ −221.46 (m, F).

EXAMPLE 7

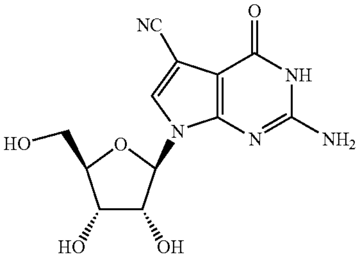
2-Amino-3,4-dihydro-4-oxo-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide



This compound was prepared following the procedures described in *Tetrahedron. Lett*. 25: 4793 (1983).

EXAMPLE 8

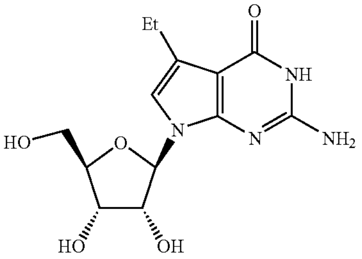
2-Amino-3,4-dihydro-4-oxo-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile



This compound was prepared following the procedures described in *J. Am. Chem. Soc*. 98: 7870 (1976).

EXAMPLE 9

2-Amino-5-ethyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



Step A: 2-Amino-7-(5-O-tert-butyldimethylsilyl-2,3-O-isopropylidene-β-D-ribofuranosyl)-4-chloro-5H-pyrrolo[2,3-d]pyrimidine

To a stirred suspension of 2-amino-4-chloro-5-ethyl-1H-pyrrolo[2,3-d]pyrimidine [described in EP 866070 (1998)] (1.57 g, 8 mmol) in dry MeCN (48 mL) was added NaH (60% in mineral oil; 0.32 g, 8 mmol), and the mixture was stirred at room temperature for 1 h. A solution of 5-O-tert-butyldimethylsilyl-2,3-O-isopropylidene-α-D-ribofuranosyl chloride [generated in situ from the corresponding lactol (1.95 g, 6.4 mmol) according to Wilcox et al., *Tetrahedron Lett*., 27: 1011 (1986)] in dry THF (9.6 mL) was added at room temperature, and the mixture was stirred overnight, then evaporated to dryness. The residue was suspended in water (100 mL) and extracted with EtOAc (200+150 mL). The combined extracts were washed with brine, dried (Na_2SO_4) and evaporated. The residue was purified on a silica gel column using a solvent system of hexanes/EtOAc: 7/1. Appropriate fractions were collected and evaporated to dryness to give the title compound (1.4 g) as a colorless foam.

Step B: 2-Amino-4-chloro-5-ethyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

A mixture of the compound from Step A (1.19 g, 2.5 mmol) in MeOH (100 mL) and water (50 mL) was stirred with DOWEX H^+ (to adjust pH of the mixture to 5) at room temperature for 2.5 h. The mixture was filtered and the resin thoroughly washed with MeOH. The combined filtrate and washings were evaporated and the residue coevaporated several times with water to yield the title compound (0.53 g) as a white solid.

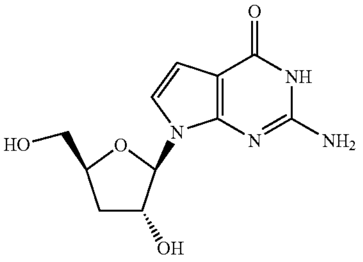
Step C: 2-Amino-5-ethyl-7-(β-D-ribofuranosyl)-7H-pyrollo[2,3-d]pyrimidin-4(3H)-one

A mixture of the compound from Step B (104 mg, 0.32 mmol) in 2N aqueous NaOH (10 mL) was stirred at reflux temperature for 15 min. The solution was cooled in ice bath, neutralized with 2 N aqueous HCl, and evaporated to dryness. The residue was suspended in MeOH, mixed with silica gel, and evaporated. The solid residue was placed onto a silica gel column (packed in a solvent mixture of CH_2Cl_2/MeOH: 10/1) which was eluted with a solvent system of CH_2Cl_2MeOH: 10/1 and 5/1. The fractions containing the product were collected and evaporated to dryness to yield the title compound (48 mg) as a white solid.

^1H NMR (CD_3OD): δ 1.22 (t, 3H), 2.69 (q, 2H), 3.69, 3.80 (2m, 2H), 4.00 (m, 1H), 4.22 (m, 1H), 4.45 (t, 1H), 5.86 (d, 1H, J=6.0 Hz), 6.60 (d, 1H, J=1.2 Hz).

EXAMPLE 10

2-Amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



Step A: 2-Amino-7-(2,3-anhydro-β-D-ribofuranosyl)-4-methoxy-7H-pyrrolo[2,3-d]pyrimidine

To a mixture of 2-amino-7-(β-D-ribofuranosyl)-4-chloro-7H-pyrrolo[2,3-d]pyrimidine (1.8 g, 6.0 mmol) in acetonitrile (80 mL) were added a solution of H_2O/CH_3CN (1:9, 1.08 mL) and then a-acetoxyisobutyryl bromide (3.5 mL, 24 mmol). After 2 h stirring at room temperature, saturated aqueous $NaHCO_3$ (170 mL) was added and the mixture was extracted with EtOAc (300+200 mL). The combined organic phase was washed with brine (100 mL), dried (Na_2SO_4) and evaporated to a pale yellow foamy residue. This was suspended in anhydrous MeOH (80 mL) and stirred overnight with 25 mL of DOWEX OH^- resin (previously washed with anhydrous MeOH). The resin was filtered, washed thoroughly with MeOH and the combined filtrate evaporated to give a pale yellow foam (1.92 g).

Step B: 2-Amino-7-(3-deoxy-β-D-ribofuranosyl)-4methoxy-7H-pyrrolo[2,3-d]pyrimidine

A solution of $LiEt_3BH$/THF (1M, 75 mL, 75 mmol) was added dropwise to a cold (ice bath) deoxygenated (Ar, 15 min) solution of the compound from Step A (1.92 g) under Ar. Stirring at 0° C. was continued for 4 h. At this point the reaction mixture was acidified with 5% aqueous acetic acid (110 mL), then purged with Ar for 1 h and and finally evaporated to a solid residue. Purification on a silica gel column using MeOH/CH_2Cl_2 as eluent yielded target compound as a colourless foam (1.01 g).

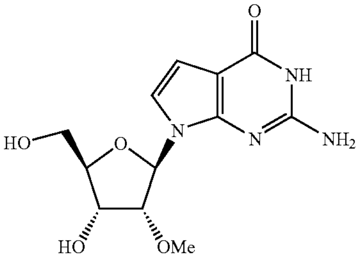
Step C: 2-Amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-4(3H)-one

A mixture of compound from Step B (0.4 g, 1.4 mmol) in 2N aqueous NaOH (40 mL) was stirred at reflux temperature for 3 h. The solution was cooled in ice bath, neutralized with 2 N aqueous HCl and evaporated to dryness. The residue was suspended in MeOH, mixed with silica and evaporated. The residue was placed onto a silica gel column which was eluted with CH_2Cl_2/MeOH: 10/1 and 5/1 to give the title compound as white solid (0.3 g).

^1H NMR (DMSO-d_6): δ 1.85, 2.12 (2m, 2H), 3.55, 3.46 (2dd, 2H), 4.18 (m, 1H); 4.29 (m, 1H), 4.85 (7, 1H), 5.42 (d, 1H) 5.82 (d, 1H, J=2.4 Hz), 6.19 (s, 2H), 6.23 (d, 1H, J=3.6 Hz), 6.87 (d, 1H), 10.31 (s, 1H).

EXAMPLE 11

2-Amino-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



Step A: 2-Amino-4-chloro-7-(5-t-butyldimethylsilyl-2,3-O-isopropylidene-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

HMPT (10.65 ml, 55 mmol) was added portionwise over 30 min. to a solution of 5-O-tert-butyldimethylsilyl-2,3-O-isopropylidene-D-ribofuranose (13.3 g, 44 mmol), dry THF (135 mL), CCl_4 (5.62 mL, 58 mmol) under N_2 at −76° C. After 30 min., the temp. was raised to −20° C. In a separate flask, a suspension of 2-amino-4-chloro-1H-pyrrolo-[2,3-d]-pyrimidine (15 g, 89 mmol) in CH_3CN (900 mL) was treated at 15° C. with 60% NaH (3.60 g., 90 mmol.). The reaction was stirred 30 min. whereupon the previous reaction mixture was cannulated with vigorous stirring. The reaction was stirred 16 hrs. and then concentrated in vacuo. The resulting semisolid was added to ice/water/EtOAc and extracted with EtOAc (3×200 mL), dried $NaSO_4$, filtered and evaporated. The resulting oil was chromatographed on silica gel (EtOAc/Hexane 1/1) to afford the product as an oil (9.0 g).

Step B: 2-Amino-4-chloro-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

A solution of the compound from Step A (5.76 g, 13 mmol) in MeOH/H_2O (1200 mL/600 mL) and Dowex WX8-400 (4.8 g) was stirred 16 hrs. at room temperature. The resin was filtered off and the filtrate evaporated to afford the title compound as a white solid; yield 3.47 g.

^1H NMR (DMSO-d_6): δ 3.56 (m, 2H), 3.86 (m, 1H), 4.07 (m, 1H), 4.32 (m, 1H), 4.99 (t, 1H), 5.10 (d, 1H), 5.30 (d, 1H), 6.00 (d, 1H), 6.38 (d, 1H), 6.71 (s br, 2H), 7.39 (d, 1H).

Step C: 2-Amino-4-chloro-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

A solution of the compound from Step B1(1.0 g, 3.3 mmol) in dry DMF (100 mL) at 15° C. was treated with 60% NaH (0.14 g, 3.5 mmol). After 30 min., iodomethane (47 g, 3.3 mmol) was added portionwise to the stirred solution. The reaction was stirred at room temperature for 16 hrs. and then evaporated at a temperature below 40° C. The resulting solid was chromatographed on silica gel to afford the product as a white solid; yield 0.81 g.

^1H NMR (DMSO-d_6): δ 3.25 (s, 3H), 3.54 (m, 2H), 3.87 (m, 1H), 4.07 (m, 1H), 4.22 (m, 1H), 5.01 (m, 1H), 5.16 (d, 1H), 6.07 (d, 1H), 6.37 (d, 1H), 6.70 (s br, 2H), 7.40 (s, 1H). Mass spectrum: m/z 316 $(M+1)^+$.

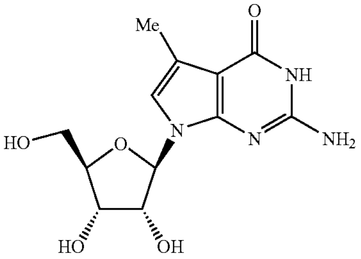
Step D: 2-Amino-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one

A solution of the compound from Step C (80 mg, 0.25 mmol) in NaOH/H_2O (1.6 g/20 ml) was heated at reflux for 7 hrs., whereupon the solution was adjusted with dilute HCl to a pH of 7 and then evaporated. Chromatography of the resulting solid on silica gel with EtOAc/MeOH 8/2 afforded the product as a white solid; yield 64 mg.

^1H NMR (DMSO-d_6): δ 3.25 (s, 3H), 3.52 (m, 2H) 3.81 (m, 1H), 4.00 (m, 1H), 4.19 (m, 1H), 5.10 (s br, 2H), 5.95 (d, 1H), 6.27 (d, 1H), 6.33 (s br, 2H), 6.95 (d, 1H), 10.55 (s br, 1H).

EXAMPLE 12

2-Amino-5-methyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



This compound is described in *Biochemistry*, 33: 2703 (1994) and was synthesized by the following procedure:

Step A: 2-Amino-7-(5-O-tert-butyldimethylsilyl-2,3-O-isopropylidene-β-D-ribofuranosyl)-4-chloro-5-methyl-7H-pyrrolo[2,3-d]pyrimidine

To a stirred suspension of 2-amino-4-chloro-5-methyl-1H-pyrrolo[2,3-d]pyrimidine (*Liebigs Ann. Chem.* 1984, 4, 708) (0.91 g, 5 mmol) in dry MeCN (30 ml) was added NaH (60% in mineral oil; 0.2 g, 5 mmol) and the mixture was stirred at room temperature for 0.5 h. A solution of 5-O-tert-butyldimethylsilyl-2,3-O-isopropylidene-α-D-ribofuranosyl chloride [generated in situ from the corresponding lactol (1.22 g, 4 mmol) according to *Tetrahedron Lett.* 27: 1011 (1986)] in dry THF (6 mL) was added at room temperature, and the mixture was stirred overnight, then evaporated to dryness. The residue was suspended in water (100 mL) and extracted with EtOAc (2×100 mL). The combined organic extracts were washed with brine, dried (Na_2SO_4) and evaporated. The residue was purified on a silica gel column using a solvent system of hexanes/EtOAc:

7/1 and 5/1. Appropriate fractions were collected and evaporated to dryness to give the title compound (0.7 g) as a colorless foam.

Step B: 2-Amino-4-chloro-5-methyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

A mixture of the intermediate from Step A (0.67 g, 1.4 mmol) in MeOH (70 ml) and water (35 ml) was stirred with DOWEX H^+ (to adjust pH of the mixture to 5) at room temperature for 4 h. The mixture was filtered and the resin thoroughly washed with MeOH. The combined filtrate and washings were evaporated and the residue coevaporated several times with water to yield the title compound (0.37 g) as a white solid.

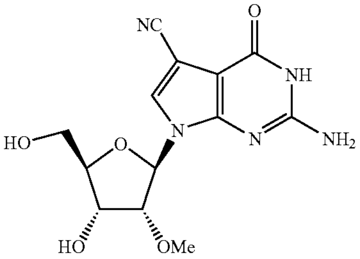
Step C: 2-Amino-5-methyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one

A mixture of intermediate from Step B (100 mg, 0.32 mmol) in 2N aqueous NaOH (20 mL) was stirred at reflux temperature for 1.5 h. The solution was cooled in ice bath, neutralized with 2 N aqueous HCl and evaporated to dryness. The residue was suspended in MeOH, mixed with silica gel and evaporated. The solid residue was placed onto a silica gel column (packed in a solvent mixture of CH_2Cl_2/MeOH: 10/1) which was eluted with a solvent system of CH_2Cl_2/MeOH: 10/1 and 5/1. The fractions containing the product were collected and evaporated to dryness to yield the title compound (90 mg) as a white solid.

^1H NMR (DMSO-d_6): δ 2.15 (d, 3H), 3.47, 3.50 (2m, 2H), 3.75 (m, 1H), 3.97 (m, 1H), 4.17 (m, 1H), 4.89 (t, 1H), 4.96 (d, 1H), 5.14 (d, 1H), 5.80 (d, 1H, J=6.4 Hz), 6.14 (s, 2H), 6.60 (q, 1H, J=1.2 Hz), 10.23 (s, 1H).

EXAMPLE 13

2-Amino-3,4-dihydro-4-oxo-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile



Step A: 2-Amino-4-chloro-7-β-D-ribofuranosyl-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile

This intermediate was prepared according to *J. Chem. Soc. Perkin Trans.* 1. 2375 (1989).

Step B: 2-Amino-4-chloro-7-[3,5-O-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile

To a solution of the compound from Step A (1.64 g, 5.00 mmol) in DMF (30 mL) was added imidazole (0.681 g, 10.0 mmol). The solution was cooled to 0° C. and 1,3-dichloro-1,1,3,3-tetraisopropyldisiloxane (1.58 g, 5.00 mmol) was added dropwise. The bath was removed and the solution stirred at room temperature for 30 minutes, evaporated in vacuo to an oil, taken up in ethyl acetate (150 mL) and washed with saturated aqueous sodium bicarbonate (50 mL) and with water (50 mL). The organic phase was dried over magnesium sulfate, filtered and evaporated in vacuo. The residue was purified on silica gel using ethyl acetate/hexane (1:2) as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (2.05 g) as a colorless foam.

^1H NMR (DMSO-d_6): δ 1.03 (m, 28H), 3.92 (m, 1H), 4.01 (m, 1H), 4.12 (m, 1H), 4.24 (m, 2H), 5.67 (m, 1H), 5.89 (s, 1H), 7.17 (bs, 2H), 8.04 (s, 1H).

Step C: 2-Amino-4-chloro-7-[2-O-methyl-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile

To a pre-cooled solution (0° C.) of the compound from Step B (1.70 g, 3.00 mmol) in DMF (30 mL) was added methyl iodide (426 mg, 3.00 mmol) and then NaH (60% in mineral oil) (120 mg, 3.00 mmol). The mixture was stirred at rt for 30 minutes and then poured into a stirred mixture of saturated aqueous ammonium chloride (100 mL) and ethyl acetate (100 mL). The organic phase was washed with water (100 mL), dried over magnesium sulfate, filtered and evaporated in vacuo. The resulting oily residue was co-evaporated three times from acetonitrile (10 mL), taken up in THF (50 mL) and tetrabutylammonium fluoride (1.1 mmol/g on silica) (4.45 g, 6.00 mmol) was added. The mixture was stirred for 30 minutes, filtered and the filtrate evaporated in vacuo. The crude product was purified on silica using methanoldichloromethane (7:93) as eluenit. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (359 mg) as a colorless solid.

^1H NMR (DMSO-d_6): δ 3.30 (s, 3H), 3.56 (m, 2H) 3.91 (m, 1H), 4.08 (m, 1H), 4.23 (m, 1H), 5.11 (m, 1H), 5.23 (m, 1H), 7.06 (m, 1H), 7.16 (bs, 2H), 8.38 (s, 1H).

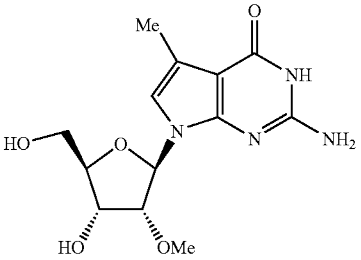
Step D: 2-Amino-3,4-dihydro-4-oxo-7-[2-O-methyl-β-D-ribofuranosyl]-7H-pyrollo[2,3-d]pyrimidine-5-carbonitrile

To a solution of the compound from Step D in DMF (5.0 mL) and dioxane (3.5 mL) was added syn-pyridinealdoxime (336 mg, 2.75 mmol) and then tetramethylguanidine (288 mg, 2.50 mmol). The resulting solution was stirred overnight at rt, evaporated in vacuo and and co-evaporated three times from acetonitrile (20 mL). The oily residue was purified on silica gel using methanol/dichloromethane (7:93) as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (103 mg) as a colorless solid.

^1H NMR (DMSO-d_6): δ 3.30 (s, 3H), 3.57 (m, 2H), 3.86 (m, 1H), 4.00 (m, 1H), 4.21 (m, 1H), 5.07 (m, 1H), 5.17 (m, 1H), 5.94 (m, 1H), 6.56 (bs, 2H), 7.93 (s, 1H), 10.82 (bs, 1H).

EXAMPLE 14

2-Amino-5-methyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



Step A: 2-Amino-4-chloro-5-methyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]-pyrimidine

Into a solution of the compound from Example 12, Step B (188 mg, 0.6 mmol) in anhydrous DMF (6 mL) was added NaH (60% in mineral oil; 26 mg, 0.66 mmol). The mixture was stirred at room temperature for 0.5 h and then cooled.

MeI (45 µL) was added at 0° C. and the reaction mixture allowed to warm to 15° C. in 5 h. Then the mixture was poured into ice-water (20 mL) and extracted with CH_2Cl_2 (100+50 mL). The combined organic extracts were washed with water (50 mL), brine (50 mL) and dried (Na_2SO_4). The evaporated residue was purified on a silica gel column with a solvent system of CH_2Cl_2/MeOH: 30/1. Appropriate fractions were pooled and evaporated to yield the title compound (50 mg) as a colorless glass.

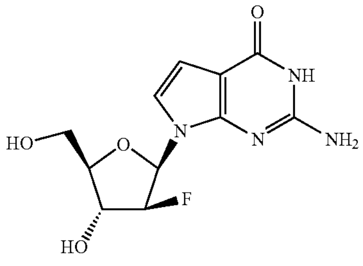
Step B: 2-Amino-7-(2-O-methyl-β-D-ribofuranosyl)-5-methyl-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one

A solution of the compound from Step A (50 mg, 0.15 mmol) in 0.5M NaOMe/MeOH (4 mL) was stirred at reflux temperature for 1.5 h. The mixture was cooled, mixed with silica gel and evaporated to dryness. The silica gel was loaded onto a silica gel column and eluted with a solvent system of CH_2Cl_2/MeOH: 30/1. The fractions containing the product were collected and evaporated to yield 2-amino-7-(2-O-methyl-β-D-ribofuranosyl)-4-methoxy-5-methyl-7H-pyrrolo[2,3-d]pyrimidine (40 mg). This was mixed with 2 N aqueous NaOH (4 mL) and stirred at reflux temperature for 10 h. The mixture was cooled in ice bath, neutralized with 2 N aqueous HCl and evaporated. The solid residue was suspended in MeOH, mixed with silica gel and evaporated. The silica gel was loaded onto a silica gel column and eluted with a solvent system of CH_2Cl_2/MeOH: 5/1. Appropriate fractions were pooled and evaporated to give the title compound (40 mg) as a white solid.

^{1}H NMR (DMSO-d_6): δ 2.18 (s, 3H), 3.26 (s, 3H), 3.45, 3.52 (2m, 2H), 3.82 (m, 1H), 3.97 (dd, 1H), 4.20 (m, 1H), 4.99 ((t, 1H), 5.10 (d, 1H), 5.94 (d, 1H, J=7.0 Hz), 6.19 (bs, 2H), 6.68 (s, 1H), 10.60 (br, 1H).

EXAMPLE 15

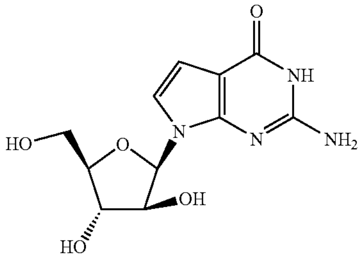
2-Amino-7-(2-deoxy-2-fluoro-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



This compound was prepared following the procedures described in *J. Med. Chem.* 38: 3957 (1995).

EXAMPLE 16

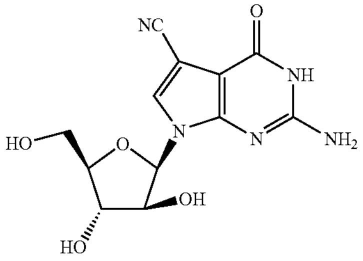
2-Amino-7-(β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



This compound was prepared following the procedures described in *J. Org. Chem.* 47: 226 (1982).

EXAMPLE 17

2-Amino-7-(β-D-arabinofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile



Step A: 2-Amino-7-(β-D-arabinofuranosyl)-4-chloro-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile

This intermediate was prepared according to *J. Chem. Soc. Perkin Trans.* 1, 2375 (1989).

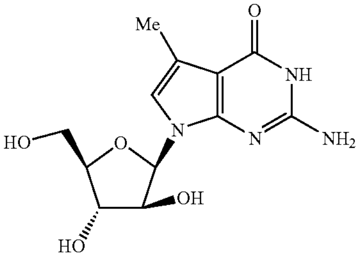
Step B: 2-Amino-7-(β-D-arabinofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile

To a solution of the compound from Step A (163 mg, 0.50 mmol) in DMF (5.0 mL) and dioxane (3.5 mL) was added syn-pyridinealdoxime (336 mg, 2.75 mmol) and then tetramethylguanidine (288 mg, 2.50 mmol). The resulting solution was stirred overnight at rt, evaporated in vacuo and and co-evaporated three times from acetonitrile (20 mL). The oily residue was purified on silica using methanol/dichloromethane (1:4) as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (72 mg) as a colorless solid.

^{1}H NMR (DMSO-d_6): δ 3.60 (m, 2H), 3.73 (m, 1H), 4.01 (m, 2H), 5.06 (m, 1H), 5.48 (m, 2H), 6.12 (m, 1H), 6.52 (bs, 2H), 7.70 (s, 1H), 10.75 (bs, 1H).

EXAMPLE 18

2-Amino-5-methyl-7-(β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



Step A 2-Amino-7-(2,3,5-tri-O-benzyl-β-D-arabinofuranosyl)-4-chloro-5-methyl-7H-pyrrolo[2,3-d]pyrimidine

To a solution of 1-O-p-nitrobenzyl-D-arabinofuranose (3.81 g, 6.70 mmol) in DCM was bubbled HBr until TLC (hexane/ethylacetate (2:1)) showed complete reaction (about 30 min). The reaction mixture was filtered and evaporated in vacuo. The oily residue was taken up in acetonitrile (10 mL) and added to a vigorously stirred suspension of 2-amino-4-chloro-5-methyl-7H-pyrrolo[2,3-d]pyrimidine (*Liebigs Ann. Chem.* (1984), 4,708) (1.11 g, 6.00 mmol) KOH (1.12 g, 20.0 mmol) and tris[2-(2-methoxyethoxy)ethyl]amine (0.216 g, 0.67 mmol) in acetonitrile (80 mL). The resulting suspension was stirred at rt for 30 min, filtered and evaporated in vacuo. The crude product was purified on silica using hexane/ethylacetate (3:1) as the eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (1.13 g) as a colorless foam.

Step B: 2-Amino-7-β-D-arabinofuranosyl-4-chloro-5-methyl-7H-pyrrolo[2,3-d]pyrimidine

To a precooled (−78° C.) solution of the compound from Step A (0.99 g, 1.7 mmol) in dichloromethane (30 mL) was added borontrichloride (1M in dichloromethane) (17 mL, 17.0 mmol) over a 10 min. The resulting solution was stirred at −78° C. for 1 h, allowed to warm to −15° C. and stirred for another 3 h. The reaction was quenched by addition of methanol/dichloromethane (1:1) (15 mL), stirred at −15° C. for 30 min, and pH adjusted to 7.0 by addition of NH_4OH. The mixture was evaporated in vacuo and the resulting oil purified on silica using methanol/dichloromethane (1:9) as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (257 mg) as a colorless foam.

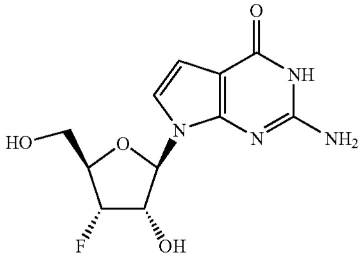
Step C: 2-Amino-7-(β-D-arabinofuranosyl)-5-methyl-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one

To the compound from Step B (157 mg, 0.50 mmol) was added NaOH (2M, aqueous) (2 mL). The resulting solution was stirred at reflux for 1 h, cooled and neutralized by addition of HCl (2M, aqueous). The mixture was evaporated in vacuo and the crude product purified on silica using methanol/dichloromethane (2:8) as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (53 mg) as a colorless powder.

^1H NMR (DMSO-d_6): δ 2.13 (d, 3H), 3.58 (m, 2H), 3.71 (m, 1H), 4.00 (m, 2H), 5.09 (m, 1H), 6.22 (bs, 2H), 5.50 (m, 2H), 6.12 (m, 1H), 6.64 (s, 1H), 10.75 (bs, 1H).

EXAMPLE 19

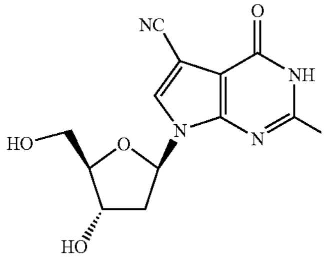
2-Amino-7-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



A solution 1-O-acetyl-2-O-benzyl-5-O-(p-toluoyl)-3-deoxy-3-fluoro-D-ribofuranose (410 mg, 1.01 mmol) (prepared by a modified method described for similar sugar derivatives, *Helv. Chim. Acta* 82: 2052 (1999) and *J. Med. Chem.*1991, 34, 2195) in anhydrous CH_2Cl_2 (1.5 mL) was cooled to −15° C. in a dry ice/CH_3CN bath. After cooling the reaction mixture for 10 min. under the argon atmosphere, 33% HBr/AcOH (370 μL, 1.5 equiv.) was added slowly over 20 min keeping the bath temperature around −15° C. After the addition was complete, the reaction mixture was stirred at −10° C. for 1 hr. The solvent was removed under reduced pressure and the residue azeotroped with anhydrous toluene (5×10 mL). In a separate flask, 2-amino-4-chloro-7H-pyrrolo[2,3-d]pyrimidine (210 mg, 1.2 mmol) was suspended in anhydrous CH_3CN (10 mL) and cooled to −10° C. To this was added 60% NaH dispersion in oil (57 mg) in two portions, and the reaction mixture was stirred for 45 min. during which time the solid dissolved and the bath temperature rose to 0° C. The bath was removed and stirring was continued for about 20 additional min. It was cooled back to −10° C. and the bromo sugar, prepared above, was taken up in anhydrous CH_3CN (1.5 mL) and added slowly to the anion of nucleobase . After the addition was complete, the reaction mixture was stirred for an additional 45 min allowing the temperature of the reaction to rise to 0° C. The bath was removed and the reaction allowed to stir at room temperature for 3 hr. Methanol was added carefully to the reaction mixture and the separated solid removed by filtration. The solvent was removed under reduced pressure and the residual oil dissolved in EtOAc (50 mL) and washed with water (3×20 mL). The organic layer was dried over Na_2SO_4 and concentrated to give an oil. It was purified by column chromatography to furnish fully protected 2-amino-7-(5-O-(p-toluoyl)-2-O-benzyl-3-deoxy-3-fluoro-β-D-ribofuranosyl)-4-chloro-7H-pyrrolo[2,3-d]pyrimidine (190 mg) as an α/β mixture (1:1). After conversion of 4 chloro to 4-oxo by heating the compound with 2N NaOH/dioxane mixture at 105° C. and after the usual workup the residue was debenzylated using 20 mol % w/w of 10% Pd/C and ammonium formate in refluxing methanol to give title compound after purification by HPLC; yield 10%. ESMS: calcd. for $C_{11}H_{13}FN_4O_4$ 284.24, found 283.0 (M+1).

EXAMPLE 20

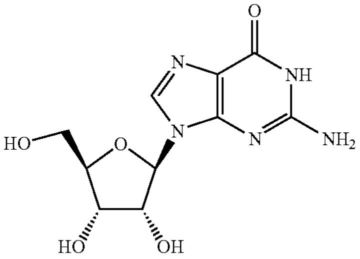
2-Amino-3,4-dihydro-4-oxo-7-(2-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile



This compound was prepared following the procedures described in *Synthesis* 1327 (1998).

EXAMPLE 21

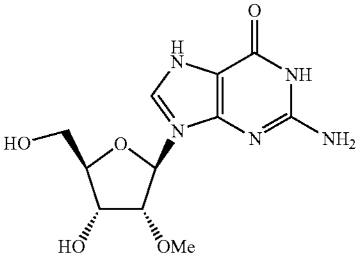
6-Amino-1-(β-D-ribofuranosyl)-1H-imidazo[4,5-d] pyridin-4(5H)-one



This compound was prepared following the conditions described in *J. Am. Chem. Soc*. 97: 2916 (1975).

EXAMPLE 22

2-Amino-7-(2-O-methyl-β-D-ribofuranosyl)-5H-pyrrolo[3,2-d]pyrimidin-4-(3H)-one

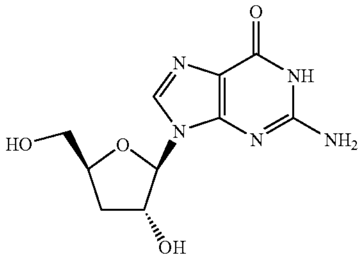


To a suspension of 2-amino-5H-pyrrolo[3,2-d]pyrimidin-4(3H)-one (9-deazaguanine) (0.454 g, 3.0 mmol) (prepared according to *J. Org. Chem*.1978, 43, 2536) and 2-O-methyl-1,3,5-tri-O-benzoyl-β-D-ribofuranose (1.54 g, 3.2 mmol) in dry nitromethane (23 mL) at 60° C. was added stannic chloride (0.54 mL, 4.5 mmol). The reaction mixture was maintained at this temperature for 0.5 hr., cooled and poured onto ice-cold saturated sodium bicarbonate solution (70 mL). The insoluble material was filtered through florisil and washed with ethyl acetate (3×50 mL). The filtrate was extracted with ethyl acetate (2×50 mL), and organic layer was washed with water (2×50 mL), dried over Na_2SO_4 and evaporated to dryness. Chromatography of the resulting foam on silica gel with CH_2Cl_2/MeOH(14:1) afforded the benzoylated product (0.419 g, 30% yield). To a suspension of the benzoylated product (0.25 g) in MeOH (2.4 mL) was added t-butylamine (0.52 mL) and stirring at room temperature was continued for 24 hrs. followed by addition of more t-butylamine (0.2 mL). The reaction mixture was stirred at ambient temperature overnight, concentrated in vacuum and the residue was purified by flash chromatography over silica gel using CH_2Cl_2/MeOH (85:15) as eluent giving the desired compound as a foam (0.80 g).

^1H NMR (200 Mz, DMSO-d_6): δ Hz3.28 (s, 3H), 3.40–3.52 (m, 3H), 3.87–3.90 (m, 1H), 4.08–4.09 (m, 1H), 4.67 (d, 1H, J=5.2 Hz), 4.74 (d, 1H, J=7.0 Hz), 5.62 and 5.50 (2 bs, 3H), 7.14 (d, 1H, J=2.6 Hz), 10.43 (s, 1H), 11.38 (s, 1H); Mass spectrum: calcd. for $C_{12}H_{16}N_4O_5$: 296.28; found: 295.11.

EXAMPLE 23

6-Amino-1-(3-deoxy-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridine-4(5H)-one (3'-deoxy-3-deaza-guanosine)



Step A: β-Deoxy-4-O-p-toluoyl-2-O-acetyl-(β-D-ribofuranosyl acetate

A solution of 3-deoxy-4-O-p-toluoyl-1,2-O-isopropylidene-β-D-ribofuranose (*Nucleosides Nucleotides* 1994, 13, 1425 and *Nucleosides Nucleotides* 1992, 11, 787) (5.85 g, 20 mmol) in 64 mL of 80% acetic acid was stirred at 85° C. overnight. The reaction mixture was concentrated and co-evaporated with toluene. The residue was dissolved in 90 mL of pyridine. Acetic anhydride (6 mL) was added at 0° C., and the reaction mixture was stirred at rt for 6 h. After condensation, the residue was dissolved in ethyl acetate and washed with aqueous sodium bicarbonate solution, water and brine. The organic phase was dried and concentrated.

Chromatographic purification on a silica gel column using 3:1 and 2:1 hexanes-EtOAc as eluent provided 5.51 g of the title compound as a clear oil.

^1H NMR ($CDCl_3$): δ 1.98 (s, 3H), 2.09 (s, 3H), 2.15–2.35 (m, 2H), 2.41 (s, 3H), 4.27–4.42 (m, 1H), 4.46–4.58 (m, 1H), 4.65–4.80 (m, 1H), 5.21–5.28 (m, 1H), 6.20 (s, 1H 7.19–7.31 (m, 2H), 7.90–8.01 (m, 2H).

Step B: Methyl 5-cyanomethyl-1-(3-deoxy4-O-p-toluoyl-2-O-acetyl-β-D-ribofuranosyl)-1H-imidazole-4-carboxylate

A mixture of methyl 5(4)-(cyanomethyl)-1H-imidazole-4(5)-carboxylate (*J. Am. Chem. Soc*. 1976, 98, 1492 and *J. Org. Chem*. 1963, 28, 3041) (1.41 g, 8.53 mmol), 1,1,1,3,3,3-hexamethyldisilazane (20.5 mL) and ammonium sulfate (41 mg) was refluxed at 125° C. under Ar atmosphere for 18 h. After evaporation, the residue was dissolved in 10 mL of dichloroethane. A solution of the compound from Step A (2.86 g, 8.5 mmol) in 10 mL of dichloroethane was added followed by addition of $SnCl_4$ (1.44 mL, 3.20 g). The resulted reaction mixture was stirred at rt overnight and diluted with chloroform. The mixture was washed with aqueous sodium bicarbonate water and brine. The organic phase was dried and concentrated. Chromatographic purification of the residue on a silica gel column using 1:1, 1:2, and 1:3 hexanes-EtOAc as eluent provided 2.06 g of the title compound as a white foam.

^{1}H NMR ($CDCl_3$) δ 2.15 (s, 3H), 2.28–2.40 (m, 2H), 2.38 (s, 3H), 3,87 (s, 3H), 4.46 (dd, 2H, J=7.6, 2.0 Hz), 4.50–4.57 (m, 1H), 4.68–4.75 (m, 1H), 4.76–4.83 (m, 1H), 5.41 (d, 1H, J=5.6 Hz), 5.91 (s, 1H), 7.24–7.28 (m, 2H), 7.80 (s, 1H), 7.82–7.90 (m, 2H); ^{13}C NMR ($CDCl_3$) δ 13.1, 20.7, 21.6, 31.5, 51.8, 63.5, 77.9, 79.2, 89.8, 115.1, 126.2, 129.3, 129.5, 131.7, 135.1, 144.3, 163.1, 166.1, 170.3.

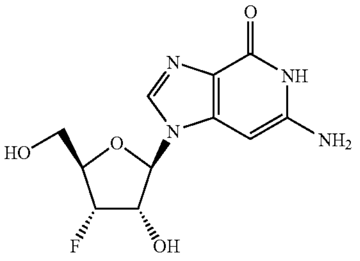
Step C: 6-Amino-1-(3-deoxy-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridine-4(5H)-one

A solution of the compound from Step B (2.00 g, 4.53 mmol) in methanol (30 mL) was saturated with ammonia at 0° C. Concentrated ammonium hydroxide (30 mL) was added and the sealed metal reactor was heated at 85° C. for 5 h. After cooling to rt, the reaction mixture was transferred directly onto a silica gel column. Elution with 4:1, 3:1 and 2:1 $CHCl_3$-MeOH provided 0.79 g of the title compound as a white solid.

^{1}H NMR (DMSO-d_6): δ 2.41–2.46 (m, 1H), 2.52–2.58 (m, 1H), 3.48–3.55 (m, 1H), 3.60–3.70 (m, 1H), 4.27–4.36 (m, 2H), 4.97 (t, 1H, J=5.6 Hz), 5.44 (s, 1H), 5.47 (s, 1H), 5.60 (s, 2H), 5.66, (d, 1H, J=4.4 Hz), 7.90 (s, 1H), 10.33 (s, 1H); ^{13}C NMR (DMSO d_6) δ 34.1, 62.4, 70.4, 74.7, 80.4, 91.6, 123.0, 136.3, 141.9, 147.6, 156.5.

EXAMPLE 24

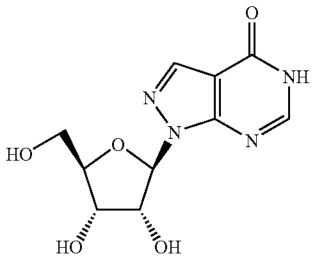
6-Amino-1-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(3H)-one



This compound was prepared in a manner similar to the preparation of 2-amino-7-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one (Example 23).

EXAMPLE 25

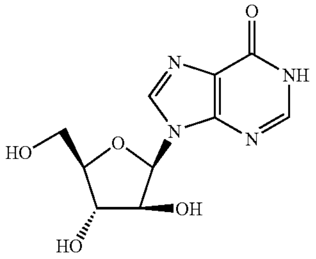
1-(β-D-Ribofuranosyl)-1H-pyrazolo[3,4-d]pyrimidin-4(3H)-one (Allopurinol riboside)



This compound was obtained from commercial sources.

EXAMPLE 26

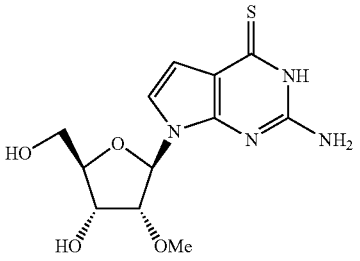
9-(β-D-Arabinofuranosyl)-9H-purin-6(1H)-one



This compound was prepared following the conditions described in *J. Med. Chem.* 18: 721(1975).

EXAMPLE 27

2-Amino-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-thione

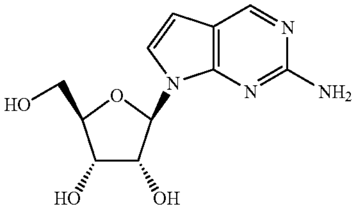


A solution of the compound from Example 11, Step C (1.5 g, 5 mmol), thiourea (0.4 g, 5.2 mmol.) in abs. EtOH was refluxed for 16 hrs. The solution was evaporated and the resulting oil chromatographed on silica gel (EtOAc/MeOH: 9/1) to afford the desired product as a foam.

^{1}H NMR (DMSO-d_6): δ 3.30 (s, 3H), 5.00–5.06 (t, 1H), 5.19 (d, 1H), 5.95 (d, 1H), 6.43 (d, 1H), (d, 1H).

EXAMPLE 28

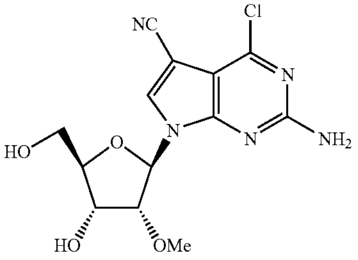
2-Amino-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



This compound was obtained from commercial sources.

EXAMPLE 29

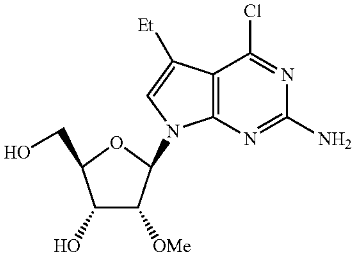
2-Amino-4-chloro-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile



This compound was prepared as described in Example 13, Steps A–C.

EXAMPLE 30

2-Amino-4-chloro-5-ethyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



Step A: 2-Amino-4-chloro-5-ethyl-7-[3,5-O-(tetraisopropyldisiloxane-1,3-diyl)-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine

To a solution of 2-amino-4-chloro-5-ethyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine (0.300 g, 0.913 mmol) in pyridine, (8 mL) was added 1,3-dichloro-1,1,3,3-tetraisopropyldisiloxane (0.317 g, 1.003 mmol) dropwise. The solution stirred at rt overnight, evaporated in vacuo to an oil, and evaporated repeatedly from acetonitrile. The crude product was purified on silica using 5% methanol in dichloromethane as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (254 mg) as a colorless solid.

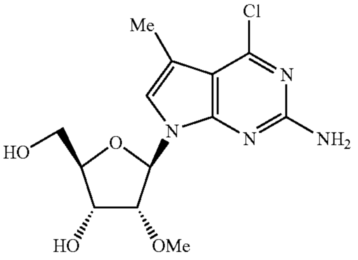
Step B: 2-Amino-4-chloro-5-ethyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

To a pre-cooled solution (0° C.) of the compound from step A (192 mg, 0.337 mmol) in DMF (3 mL) was added methyl iodide (45.4 mg, 0.320 mmol) and then NaH (60% in mineral oil) (8.10 mg, 0.320 mmol). The mixture was stirred at rt for 45 minutes and then poured into a stirred mixture of saturated aqueous ammonium chloride (10 mL) and ethyl acetate (10 mL). The organic phase phase was washed with brine (10 mL) and dried over $MgSO_4$ and evaporated in vacuo. The resulting oily residue was taken up in THF (5 mL) and tetrabutylammonium fluoride (1.1 mmol/g on silica) (0.529 g, 0.582 mmol) was added. The mixture was stirred for 30 minutes, filtered and the filtrate evaporated in vacuo. The crude product was purified on silica using 10% methanol in dichloromethane as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (66 mg) as a colorless solid.

^1H NMR (DMSO-d_6): δ 1.15 (t, 3H), 2.65 (q, 2H), 3.20 (s, 3H), 3.51 (m, 2H), 3.84 (m, 1H), 4.04 (m, 1H), 4.21 (m, 1H), 4.99 (m, 2H), 5.15 (m, 2H), 6.07 (m, 2H), 6.62 (s br, 2H), 7.06 (s, 2H).

EXAMPLE 31

2-Amino-4-chloro-5-methyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

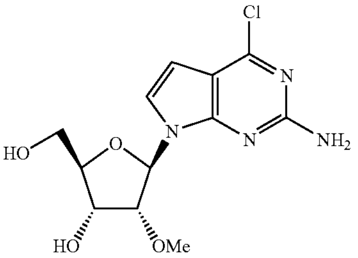


This compound was prepared as described in Example 14, Step A.

^1H NMR (CD_3OD): δ 2.33 (s, 3H), 3.39 (s, 1H), 3.72, 3.83 (2dd, 2H), 4.03 (m, 1H), 4.17 (t, 1H), 4.39 (dd, 1H), 5.98 (d, 1H, J=5.9 Hz), 6.7 (bs, 2H), 7.01 (s, 1H).

EXAMPLE 32

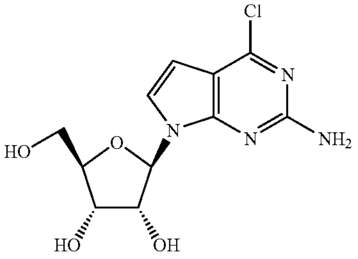
2-Amino-4-chloro-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



This compound was synthesized as described in Example 11, Steps A–C.

EXAMPLE 33

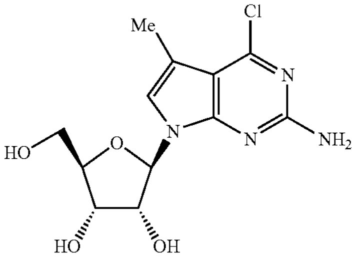
2-Amino-4-chloro-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



This compound was prepared following the procedures described in *Helv. Chim. Acta* 73: 1879 (1990).

EXAMPLE 34

2-Amino-4-chloro-5-methyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

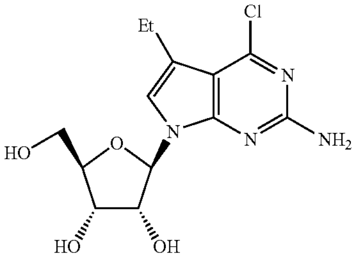


The compound was prepared as described in Example 12, Steps A–B.

^{1}H NMR (DMSO-d_6): δ 2.29 (s, 3H), 3.54 (m, 2H), 3.84 (m, 1H), 4.04 (dd, 1H, J_1=3.0, J_2=4.9 Hz), 4.80–5.50 (bs, 3H), 4.28 (t, 1H), 5.98 (d, 1H, J=6.5 Hz), 6.7 (bs, 2H), 7.13 (s, 1H).

EXAMPLE 35

2-Amino-4-chloro-5-ethyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

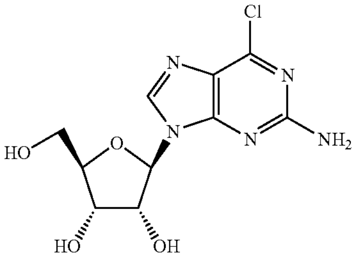


This compound was prepared as described in Example 9, Steps A–B.

^{1}H NMR (DMSO-d_6): δ 2.00 (t, 3H), 2.69 (q, 2H), 3.48 (dd, 1 H, J_1=4.2 Hz, J_2=11.8 Hz), 3.56 (dd, 1H, J_1=4.3 Hz, J_2=11.8 Hz), 3.80 (m, 1H), 4.02 (dd, 1H, J_1=3.1 Hz, J_2=5.0 Hz), 4.62 (t, 1H), 5.0 (bs, 2H), 5.2 (bs, 1H), 5.60 (d, 1H, J=6.4 Hz), 6.61 (bs, 2H), 7.09 (s, 1H).

EXAMPLE 36

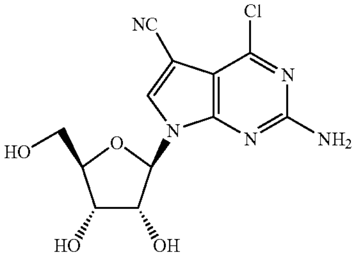
2-Amino-6-chloro-9-(β-D-ribofuranosyl)-9H-purine



This compound was obtained from commercial sources.

EXAMPLE 37

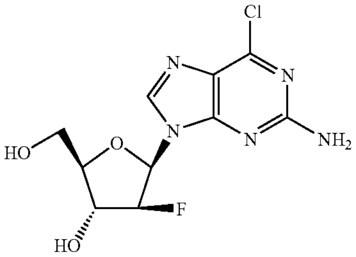
2-Amino-4-chloro-7-(β-D-ribofuranosyl)-7H-pyrollo[2,3-d]pyrimidine-5-carbonitrile



This compound was prepared following the procedures described in *J. Chem. Soc. Perkin Trans.* 1, 2375 (1989).

EXAMPLE 38

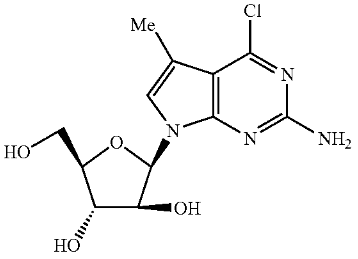
2-Amino-4-chloro-7-(2-deoxy-2-fluoro-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



This compound was prepared following the procedures described in *J. Med. Chem.* 38: 3957 (1995).

EXAMPLE 39

2-Amino-4-chloro-5-methyl-7-(β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

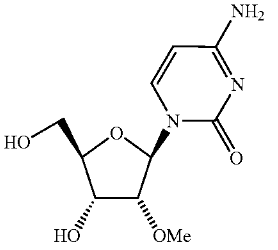


The compound was prepared as described in Example 18, Steps A–B.

^{1}H NMR (DMSO-d$_6$): δ 2.24 (s, 3H), 3.60 (m, 3H), 3.98 (m, 2H), 4.98 (m, 1H), 5.43 (bs, 2H), 6.25 (s, 1H), 6.57 (bs, 2H), 7.01 (s, 1H).

EXAMPLE 40

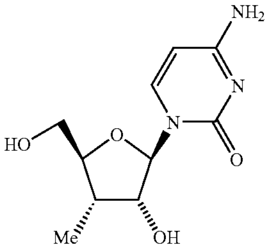
2'-O-Methylcytidine



This compound was obtained from commercial sources.

EXAMPLE 41

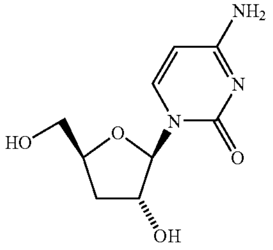
3'-Deoxy-3'-methylcytidine



This compound was prepared following the procedures described in U.S. Pat. No. 3,654,262 (1972), which is incorporated by reference herein in its entirety.

EXAMPLE 42

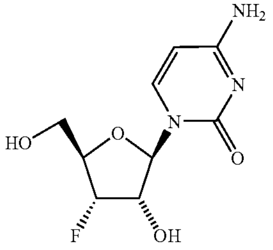
3'-Deoxycytidine



This compound was obtained from commercial sources.

EXAMPLE 43

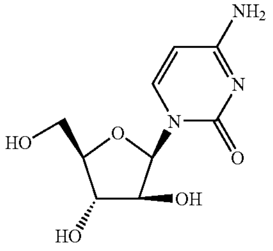
3'-Deoxy-3'-fluorocytidine



This compound was prepared following the procedures described in *J. Med. Chem.* 34: 2195 (1991).

EXAMPLE 44

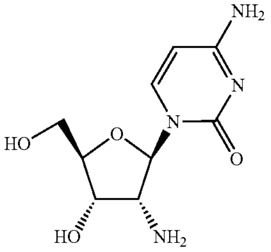
1-(β-D-Arabinofuranosyl)-1H-cytosine



This compound was obtained from commercial sources.

EXAMPLE 45

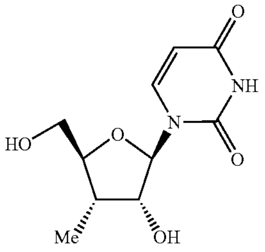
2'-Amino-2'-deoxycytidine



This compound was obtained from commercial sources.

EXAMPLE 46

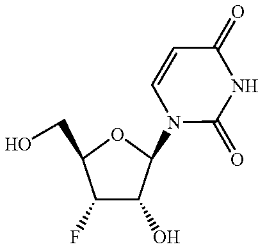
3'-Deoxy-3'-methyluridine



This compound was prepared following procedures described in U.S Pat. No. 3,654,262, which is incorporated by reference herein in its entirety.

EXAMPLE 47

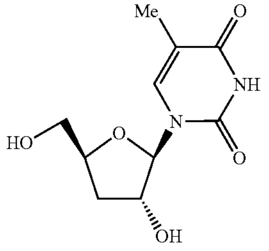
3'-Deoxy-3'-fluorouridine



This compound was prepared following procedures described in *J. Med. Chem.* 34: 2195 (1991) and *FEBS Lett.* 250: 139 (1989).

EXAMPLE 48

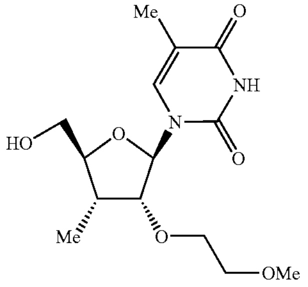
3'-Deoxy-5-methyluridine



This compound was obtained from commercial sources.

EXAMPLE 49

3'-Deoxy-2'-O-(2-methoxyethyl)-3'-methyl-5-methyluridine



Step A: 5'-O-(tert-butyldiphenylsilyl)-3'-O-(3-tert-butylphenoxythiocarbonyl)-2'-O-(2-methoxyethyl)-5-methyluridine

This compound was synthesized by the reaction of the corresponding 5'-protected-2'-substituted-5-methyluridine with 3'-t-butylphenoxy chlorothionoformate following the similar procedure for the preparation of 3'-phenoxythiocarbonyl-2'-deoxy derivative (*Synthesis* 1994, 1163).

Step B: 5'-O-(tert-Butyldiphenylsilyl)-3'-deoxy-2'-O-(2-methoxyethyl)-3'-(2-phenylethenyl)-5-methyluridine

To a solution of 5'-O-(tert-butyldiphenylsilyl)-3'-O-(3-tert-butylphenoxythiocarbonyl)-2'-O-(2-methoxyethyl)-5-methyluridine (15.0 g, 20.0 mmol) in 150 mL of benzene was added PhCH═CHSnBu$_3$ (18.7 g, 50 mmol). The resulting solution was degassed three times with argon at rt and 45° C. After AIBN (1.0 g, 6.1 mmol) was added, the resulting solution was refluxed for 2 h. Another portion of AIBN (1.0 g, 6.1 mmol) was added after cooling to about 40° C. and refluxed for 2 h. This procedure was repeated until the starting material disappeared. The solvent was evaporated and the residue was purified by flash chromatography on a silica gel column using 10:1 and 5:1 hexanes-EtOAc as eluent to give 1.74 g of 5'-O-(tert-butyldiphenylsilyl)-3'-deoxy-2'-O-(2-methoxyethyl)-3'-(2-phenylethenyl)5-methyluridine as a white foam.

^{1}H NMR (CDCl$_3$): δ 1.13, (s, 9H), 1.43 (s, 3H), 3.18–3.30 (m, 1H), 3.37 (s, 3H), 3.58–3.62 (m, 2H), 3.79–3.80 (m, 2H), 4.06–4.37 (m, 4H), 4.95 (s, 1H), 6.25–6.40 (m, 1H), 6.62 (d, 1H, J=16 Hz), 7.27–7.71 (m, 16H), 9.21 (s, 1H); ^{13}C NMR (CDCl$_3$) δ 11.9, 19.6, 27.2, 45.3, 59.0, 62.1, 70.2, 72.0, 84.6, 87.1, 90.2, 110.4, 122.8, 126.4, 127.8, 128.0, 128.3, 128.6, 130.0, 132.7, 133.5, 134.7, 135.3, 135.4, 136.9, 150.3, 154.1; HRMS (FAB) m/z 641.302 (M+H)$^+$ ($C_{37}H_{45}N_2O_6Si$ requires 641.304).

Step C: 5'-O-(tert-Butyldiphenylsilyl)-3'-deoxy-3'-(hydroxymethyl -2'-O-(2-methoxyethyl)-5-methyluridine

To a solution of 5'-O-(tert-butyldiphenylsilyl)-3'-deoxy-2'-O-(2-methoxyethyl)-3'-(2-phenylethenyl)-5-methyluridine. (5.0 g, 7.8 mmol) and N-methylmorpholine N-oxide (NMO) (1.47 g, 12.5 mmol) in 150 mL of dioxane was added a catalytic amount of osmium tetraoxide (4% aqueous solution, 2.12 mL, 85 mg, 0.33 mmol). The flask was covered by aluminum foil and the reaction mixture was stirred at rt overnight. A solution of $NaIO_4$ (5.35 g, 25 mmol) in 5 mL of water was added to the above stirred reaction mixture. The resulting reaction mixture was stirred for 1 h at 0° C. and 2 h at rt, followed by addition of 10 mL of ethyl acetate. The mixture was filtered through a celite pad and washed with ethyl acetate. The filtrate was washed 3 times with 10% aqueous $Na_2S_2O_3$ solution until the color of aqueous phase disappeared. The organic phase was further washed with water and brine, dried (Na_2SO_4) and concentrated. The aldehyde thus obtained was dissolved in 130 mL of ethanol-water (4:1, v/v). Sodium borohydride ($NaBH_4$) (1.58 g, 40 mmol) was added in portions at 0° C. The resulting reaction mixture was stirred at rt for 2 h and then treated with 200 g of ice water. The mixture was extracted with ethyl acetate. The organic phase was washed with water and brine, dried (Na_2SO_4) and concentrated. The resulted residue was purified by flash chromatography on a silica gel column using 2:1, 1:1 and 1:2 hexanes-EtOAc as eluents to give 1.6 g of 5'-O-(tert-butyldiphenylsilyl)-3'-deoxy-3'-(hydroxymethyl)-2'-O-(2-methoxyethyl)-5-methyluridine as a white foam.

^{1}H NMR ($CDCl_3$): δ 1.09 (s, 9H), 1.50 (s, 3H), 2.25 (bs, 1H), 2.52–2.78 (m, 1H), 3.38 (s, 3H), 3.52–4.25 (m, 10H), 5.86 (s, 1H), 7.38–7.70 (m, 11H), 9.95 (bs, 1H); ^{13}C NMR ($CDCl_3$): δ 12.1, 19.5, 27.1, 43.1, 58.2, 58.8, 63.1, 69.5, 71.6, 82.3, 86.1, 89.8, 110.5, 128.0, 130.2, 132.5, 133.2, 135.1, 135.3, 136.5, 150.5, 164.4; HRMS (FAB) m/z 569.268 (M+H)$^+$ ($C_{30}H_{41}N_2O_7Si$ requires 569.268).

Step D: 5'-O-(tert-Butyldiphenylsilyl -3'-deoxy-3'-(iodomethyl)-2'-O-(2-methoxyethyl)-5-methyluridine

To a solution of 5'-O-(tert-butyldiphenylsilyl)-3'-deoxy-3'-(hydroxymethyl)-2'-O-(2-methoxyethyl) -5-methyluridine (1.34 g, 2.35 mmol) in 25 mL of anhydrous DMP under stirring was added sequentially at 0° C. 2,6-lutidine (0.55 mL, 0.51 g, 4.7 mmol, 2.0 equiv) and methyl triphenoxy-phosphonium iodide (1.28 g, 2.83 mmol). The resulting reaction mixture was stirred at 0° C. for 1 h and at rt for 2 h. The reaction mixture was diluted with 10 mL of ethyl acetate and washed twice with 0.1 N $Na_2S_2O_3$ aqueous solution to remove iodine. The organic phase was further washed with aqueous $NaHCO_3$ solution, water, and brine. The aqueous phases were back extracted with ethyl acetate. The combined organic phases were dried (Na_2SO_4) and concentrated. The resulting residue was purified by flash chromatography on a silica gel column using 5:1, 3:1 and then 1:1 hexanes-EtOAc to provide 1.24 g of 5'-O-(tert-butyldiphenylsilyl)-3'-deoxy-3'-(iodomethyl)-2'-O-(2-methoxyethyl)-5-methyluridine as a white foam.

^{1}H NMR ($CDCl_3$): δ 1.13 (s, 9H), 1.62 (s, 3H), 2.64–2.85 (m, 2H), 3.20–3.35 (m, 1H), 3.38 (s, 3H), 3.50–4.25 (m, 8H), 5.91 (s, 1H), 7.32–7.50 (m, 6H), 7.60 (s, 1H), 7.62–7.78 (m, 4H), 10.46 (s, 1H); ^{13}C NMR ($CDCl_3$): δ 12.4, 19.5, 27.2, 45.0, 58.0, 62.5, 70.3, 71.9, 83.3, 85.6, 88.9, 110.5, 128.1, 128.2, 130.1, 130.3, 132.4, 132.9, 135.0, 135.4, 135.6, 150.7, 164.7; HRMS (FAB) m/z 679.172 (M+H)$^+$ ($C_{30}H_{40}IN_2O_6Si$ requires 679.170).

Step E: 3'-Deoxy-3'-tiodomethyl)-2'-O-(2-methoxyethyl)-5-methyluridine

A solution of 5'-O-(tert-butyldiphenylsilyl)-3'-deoxy-3'-(iodomethyl)-2'-O-(2-methoxyethyl)-5-methyluridine (1.12 g, 1.65 mmol) and triethylamine trihydrofluoride (1.1 mL, 1.1 g, 6.7 mmol) in 20 mL of THF was stirred at rt for 24 h. The reaction mixture was diluted with 50 mL of ethyl acetate and washed with water and brine. The organic phase was dried (Na_2SO_4) and concentrated. The residue was purified by flash chromatography on a silica gel column. Gradient elution with 2:1, 1:2 and then 1:3 hexanes-EtOAc provided 504 mg of the title compound as a white foam.

^{1}H NMR (CD_3OD): δ 1.87 (s, 3H), 2.47–2.75 (m, 1H), 3.18–3.37 (m, 2H), 3.40 (s, 3H), 3.59–3.70 (m, 2H), 3.71–3.90 (m, 2H), 3.92–4.17 (m, 4H), 5.87 (s, 1H), 8.17 (s, 1H); ^{13}C NMR (CD_3OD): δ 12.5, 45.2, 59.2, 60.9, 71.0, 72.9, 85.4, 87.3, 89.7, 110.5, 138.0, 152.1, 166.6; HRMS (FAB) m/z 441.053 (M+H)$^+$ ($C_{14}H_{22}IN_2O_6$ requires 441.052).

Step F: 3'-Deoxy-5'-O-(4-methoxytrityl)-3'-(iodomethyl)-2'-O-(2-methoxyethyl)-5-methyluridine

A mixture of 3'-deoxy-3'-(iodomethyl)-2'-O-(2-methoxyethyl)-5-methyluridine (472 mg, 1.1 mmol), diisopropylethylamine (0.79 mL, 0.586 g, 4.5 mmol), and p-anisyl chlorodiphenyl methane (4'-methoxytrityl chloride, MMT-Cl) (1.32 g, 4.27 mmol) in 6 mL of ethyl acetate and 4 mL of THF was stirred at rt for 48 h. The reaction mixture was diluted with ethyl acetate and washed with water, followed by brine. The organic phase was dried (Na_2SO_4) and concentrated. The crude product was purified by flash chromatography on a silica gel column. Gradient elution with 3:1, 2:1, 1:1, and then 1:3 hexanes-EtOAc provided 690 mg of the title compound as a white foam.

^{1}H NMR ($CDCl_3$): δ 1.46 (s, 3H), 2.70–2.89 (m, 2H), 3.19–3.31 (m, 2H), 3.39 (s, 3H), 3.58–3.70 (m, 3H), 3.80 (s, 3H), 3.80–3.94 (m, 1H), 4.05–4.25 (m, 3H), 5.89 (s, 1H), 6.85 (s, 1H), 6.89 (s, 1H), 7.24–7.48 (m, 12H), 7.78 (s, 1H), 9.69 (s, 1H); ^{13}C NMR ($CDCl_3$): δ 12.3, 45.3, 55.3, 58.9, 61.6, 70.2, 71.9, 82.6, 85.6, 87.1, 89.1, 110.5, 113.4, 127.4, 128.2, 128.4, 130.5, 134.7, 135.3, 143.6, 143.7, 150.5, 158.9, 164.6. HRMS (FAB) m/z 735.155 (M+Na)$^+$ ($C_{34}H_{37}IN_2O_7Na$ requires 735.154).

Step G: 3'-Deoxy-5'-O-(4-methoxytrityl)-3'-methyl-2'-O-(2-methoxyethyl)-5-methyluridine

A mixture of ammonium phosphinate (410 mg, 5.1 mmol) and 1,1,1,3,3,3-hexamethyldisilazane (1.18 mL, 0.90 g, 5.59 mmol) was heated at 100–110° C. for 2 h under nitrogen atmosphere with condenser. The intermediate BTSP(bis [trimethylsilyl]phosphinate) was cooled to 0° C. and 5 mL of dichloromethane was injected. To this mixture was injected a solution of 3'-deoxy-5'-O-(4-methoxytrityl)-3'-(iodomethyl)-2'-O-(2-methoxyethyl)-5-methyluridine (0.78 g, 1.1 mmol) and diisopropylethylamine (0.39 mL, 287 mg, 2.23 mmol) in 7 mL of dichloromethane. After the reaction mixture was stirred at rt overnight, a mixture of THF-MeOH-NEt_3 (3/6/0.3 mL) was added and continued to stir for 1 h. The reaction mixture was filtered through a pad of celite and washed with dichloromethane. The solvent was evaporated and the residue was purified by flash chromatography on a silica gel column using 2:1, 1:1, and then 1:2 hexanes-EtOAc as eluent providing 380 mg of the title compound.

^{1}H NMR ($CDCl_3$): δ 0.97 (d, 3H, J=6.8 Hz), 1.41 (s, 3H), 2.35–2.55 (m, 1H), 3.27 (dd, 1H, J=11.0, 3.0 Hz), 3.37 (s, 3H), 3.54–3.68 (m, 3H), 3.79 (s, 3H), 3.75–3.87 (m, 1H), 3.94 (d, 1H, J=5.0 Hz), 4.03–4.16 (m, 2H), 5.84 (s, 1H), 6.83 (s, 1H), 6.87 (s, 1H), 7.20–7.37 (m, 8H), 7.39–7.50 (m, 4H), 7.86 (s, 1H), 9.50 (s, 1H; ^{13}C NMR ($CDCl_3$): δ 8.7, 12.1, 35.6, 55.3, 59.0, 61.7, 69.8,72.1, 85.4, 86.4, 86.7, 89.8, 110.0, 113.3, 127.2, 128.0, 128.4, 130.4, 135.0, 135.7, 143.9, 150.5, 158.8, 164.6. HRMS (FAB) m/z 609.256 $(M+Na)^+$ ($C_{34}H_{38}N_2O_7Na$ requires 609.257).

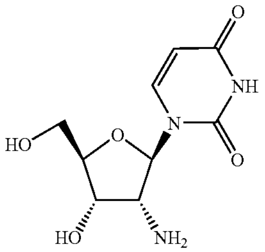
Step H: 3'-Deoxy-3'-methyl-2'-O-(2-methoxyethyl)-5-methyluridine

Trifluoroacetic acid (1.5 mL) was added dropwise to a stirred solution of 3'-deoxy-5'-O-(4-methoxytrityl)-3'-methyl-2'-O-(2-methoxyethyl)-5-methyluridine (370 mg, 0.63 mmol) in 50 mL of chloroform at 0° C. The mixture was stirred at rt for 30 min, concentrated, and then dissolved in ethyl acetate. The solution was washed with dilute sodium bicarbonate and brine. The organic phase was dried (Na_2SO_4) and concentrated. The resulting residue was purified by flash chromatography on a silica gel column. Elution with 1:1, 1:3 and then 0:1 hexanes-EtOAc provided 170 mg of the title compound as a white foam.

^1H NMR ($CDCl_3$): δ 1.03 (d, 3H, J=6.8 Hz), 1.83 (s, 3H) 2.20–2.40 (m, 1H), 3.10–3.28 (m, 1H), 3.35 (s, 3H), 3.50–4.15 (m, 10H), 5.81 (s, 1H), 7.89 (s, 1H), 9.77 (s, 1H); ^{13}C NMR ($CDCl_3$): δ 8.9 12.4, 34.7, 59.0, 60.6, 69.7,72.0, 86.3, 89.8, 109.7, 136.9, 150.4, 164.7. HRMS (FAB) m/z 315.154 $(M+H)^+$ ($C_{14}H_{23}N_2O_6$ requires 315.155).

EXAMPLE 50

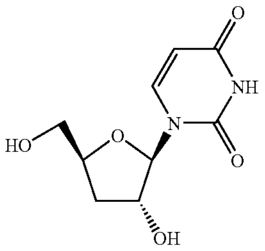
2'-Amino-2'-deoxyuridine



This compound was prepared following the procedures described in *J. Org. Chem*.61: 781 (1996).

EXAMPLE 51

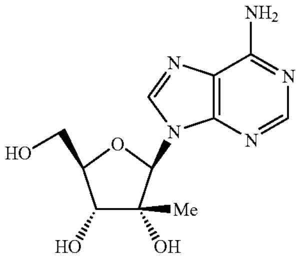
3'-Deoxyuridine



This compound was obtained from commercial sources.

EXAMPLE 52

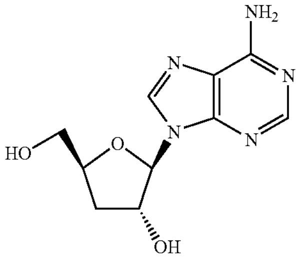
2'-C-Methyladenosine



This compound was prepared following the conditions described in *J.Med. Chem*. 41: 1708 (1998).

EXAMPLE 53

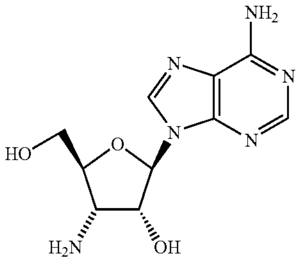
3'-Deoxyadenosine (Cordycepin)



This compound was obtained from commercial sources.

EXAMPLE 54

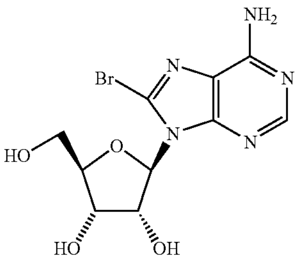
3'-Amino-3'-deoxyadenosine



This compound was prepared following the conditions described in *Tetrahedron Lett*. 30: 2329 (1989).

EXAMPLE 55

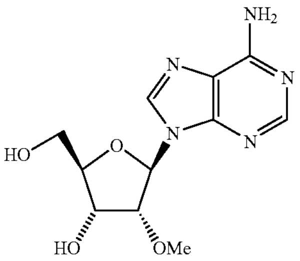
8-Bromoadenosine


NH2
N
N
Br
N
N
HO
O
HO
OH

This compound was obtained from commercial sources.

EXAMPLE 56

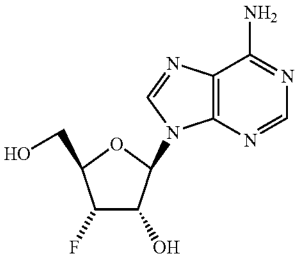
2'-O-Methyladenosine


NH2
N
N
N
N
HO
O
HO
OMe

This compound was obtained from commercial sources.

EXAMPLE 57

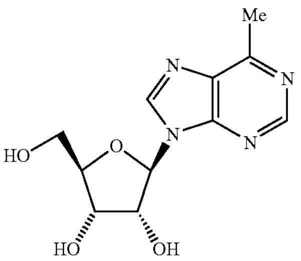
3'-Deoxy-3'-fluoroadenosine


NH2
N
N
N
N
HO
O
F
OH

This compound was prepared following the procedures described in *J. Med. Chem.* 34: 2195 (1991).

EXAMPLE 58

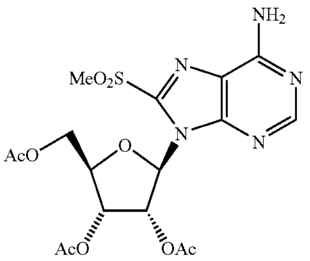
6-Methyl-9-(β-D-ribofuranosyl)-9H-purine


Me
N
N
N
N
HO
O
HO
OH

This compound was prepared following the procedures described in *Nucleosides, Nucleotides, Nucleic Acids* 19: 1123 (2000).

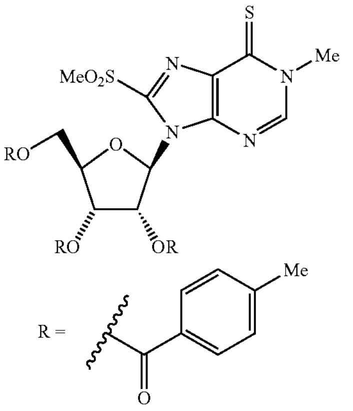
EXAMPLE 59

2',3',5'-tri-O-acetyl-8-methylsulfonyladenosine


NH2
MeO2S
N
N
N
N
AcO
O
AcO
OAc

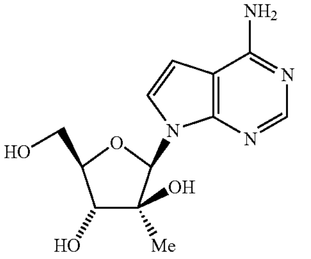
EXAMPLE 60

1-Methyl-9-[2,3,5-tri-O-(p-toluoyl)-β-D-ribofuranosyl]-9H-purine-6(1H)-thione


S
MeO2S
N
N
Me
N
N
RO
O
RO
OR
Me
R =
O

EXAMPLE 61

4-Amino-7-(2-C-methyl-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

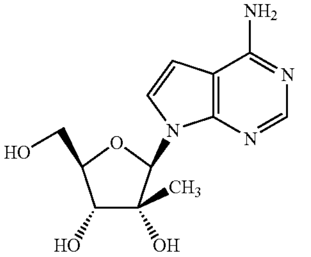


To chromium trioxide (1.57 g, 1.57 mmol) in dichloromethane (DCM) (10 mL) at 0° C. was added acetic anhydride (145 mg, 1.41 mmol) and then pyridine (245 mg, 3.10 mmol). The mixture was stirred for 15 min, then a solution of 7-[3,5-O-[1,1,3,3-tetrakis(1-methylethyl)-1,3-disiloxanediyl]-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidin-4-amine [for preparation, see *J. Am. Chem. Soc.* 105: 4059 (1983)] (508 mg, 1.00 mmol) in DCM (3 mL) was added. The resulting solution was stirred for 2 h and then poured into ethyl acetate (10 mL), and subsequently filtered through silica gel using ethyl acetate as the eluent. The combined filtrates were evaporated in vacuo, taken up in diethyl ether/TBF (1:1) (20 mL), cooled to −78° C. and methylmagnesium bromide (3M, in TEF) (3.30 mL, 10 mmol) was added dropwise. The mixture was stirred at −78° C. for 10 min, then allowed to come to room temperature (rt) and quenched by addition of saturated aqueous ammonium chloride (10 mL) and extracted with DCM (20 mL). The organic phase was evaporated in vacuo and the crude product purified on silica gel using 5% methanol in dichloromethane as eluent. Fractions containing the product were pooled and evaporated in vacuo. The resulting oil was taken up in THF (5 mL) and tetrabutylammonium fluoride (TBAF) on silica (1.1 mmol/g on silica) (156 mg) was added. The mixture was stirred at rt for 30 min, filtered, and evaporated in vacuo. The crude product was purified on silica gel using 10% methanol in dichloromethane as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired compound (49 mg) as a colorless solid.

^{1}H NMR (DMSO-d$_6$): δ 1.08 (s, 3H), 3.67 (m, 2H), 3.74 (m, 1H), 3.83 (m, 1H), 5.19 (m, 1H), 5.23 (m, 1H), 5.48 (m, 1H), 6.08 (1H, s), 6.50 (m, 1H), 6.93 (bs, 2H), 7.33 (m, 1H), 8.02 (s, 1H).

EXAMPLE 62

4-Amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



Step A: 3,5-Bis-O-(2,4-dichlorophenylmethyl)-1-O-methyl-α-D-ribofuranose

A mixture of 2-O-acetyl-3,5-bis-O-(2,4-dichlorophenylmethyl)-1-O-methyl-α-D-ribofuranose [for preparation, see: Helv. Chim. Acta 78: 486 (1995)] (52.4 g, 0.10 mol) in methanolic K_2CO_3 (500 mL, saturated at room temperature) was stirred at room temperature for 45 min. and then concentrated under reduced pressure. The oily residue was suspended in CH_2Cl_2 (500 mL), washed with water (300 mL+5×200 mL) and brine (200 mL), dried (Na_2SO_4), filtered, and concentrated to give the title compound (49.0 g) as colorless oil, which was used without further purification in Step B below.

^{1}HMR (DMSO-d$_6$): δ 3.28 (s, 3H, OCH_3), 3.53 (d, 2H, $J_{5,4}$=4.5 Hz, H-5a, H-5b), 3.72 (dd, 1H, $J_{3,4}$=3.6 Hz, $J_{3,2}$=6.6 Hz, H-3), 3.99 (ddd, 1H, $J_{2,1}$=4.5 Hz, $J_{2,OH\text{-}2}$=9.6 Hz, H-2), 4.07 (m, 1H, H-4), 4.50 (s, 2H, CH_2Ph), 4.52, 4.60 (2d, 2H, J_{gem}=13.6 Hz, CH_2Ph), 4.54 (d, 1H, OH-2), 4.75 (d, 1H, H-1), 7.32–7.45, 7.52–7.57 (2m, 10H, 2Ph).

^{13}C NMR (DMSO-d$_6$) δ 55.40, 69.05, 69.74, 71.29, 72.02, 78.41, 81.45, 103.44, 127.83, 127.95, 129.05, 129.28, 131.27, 131.30, 133.22, 133.26, 133.55, 133.67, 135.45, 135.92.

Step B: 3,5-Bis-O-(2,4-dichlorophenylmethyl)-1-O-methyl-α-D-erythro-pentofuranos-2-ulose

To an ice-cold suspension of Dess-Martin periodinane (50.0 g, 118 mmol) in anhydrous CH_2Cl_2 (350 mL) under argon (Ar) was added a solution of the compound from Step A (36.2 g, 75 mmol) in anhydrous CH_2Cl_2 (200 mL) dropwise over 0.5 h. The reaction mixture was stirred at 0° C. for 0.5 h and then at room temperature for 3 days. The mixture was diluted with anhydrous Et_2O (600 mL) and poured into an ice-cold mixture of $Na_2S_2O_3.5H_2O$ (180 g) in saturated aqueous $NaHCO_3$ (1400 mL). The layers were separated, and the organic layer was washed with saturated aqueous $NaHCO_3$ (600 mL), water (800 mL) and brine (600 mL), dried ($MgSO_4$), filtered and evaporated to give the title compound (34.2 g) as a colorless oil, which was used without further purification in Step C below.

^{1}H NMR ($CDCl_3$) δ 3.50 (s, 3H, OCH_3), 3.79 (dd, 1H, $J_{5a,5b}$=11.3 Hz, $J_{5a,4}$=3.5 Hz, H-5a), 3.94 (dd, 1H, $J_{5b,4}$=2.3 Hz, H-5b), 4.20 (dd, 1H, $J_{3,1}$=1.3 Hz, $J_{3,4}$=8.4 Hz, H-3), 4.37 (ddd, 1H, H-4), 4.58, 4.69 (2d, 2H, J_{gem}=13.0 Hz, CH_2Ph), 4.87 (d, 1H, H-1), 4.78, 5.03 (2d, 2H, J_{gem}=12.5 Hz, CH_2Ph), 7.19–7.26, 7.31–7.42 (2m, 10H, 2Ph).

^{13}C NMR (DMSO-d$_6$) δ 55.72, 69.41, 69.81, 69.98, 77.49, 78.00, 98.54, 127.99, 128.06, 129.33, 129.38, 131.36, 131.72, 133.61, 133.63, 133.85, 133.97, 134.72, 135.32, 208.21.

Step C: 3,5-Bis-O-(2,4-dichlorophenylmethyl)-2-C-methyl-1-O-methyl-α-D-ribofuranose

To a solution of MeMgBr in anhydrous Et_2O (0.48 M, 300 mL) at −55° C. was added dropwise a solution of the compound from Step B (17.40 g, 36.2 mmol) in anhydrous Et_2O (125 mL). The reaction mixture was allowed to warm to −30° C. and stirred for 7 h at −30° C. to −15° C., then poured into ice-cold water (500 mL) and the mixture vigorously stirred at room temperature for 0.5 h. The mixture was filtered through a Celite pad (10×5 cm) which was thoroughly washed with Et_2O. The organic layer was dried ($NgSO_4$), filtered and concentrated. The residue was dissolved in hexanes (~30 mL), applied onto a silica gel column (10×7 cm, prepacked in hexanes) and eluted with hexanes and hexanes/EtOAc (9/1) to give the title compound (16.7 g) as a colorless syrup.

^{1}H NMR ($CDCl_3$): δ 1.36 (d, 3H, $J_{Me,OH}$=0.9 Hz, 2C-Me), 3.33 (q, 1H, OH), 3.41 (d, 1H, $J_{3,4}$=3.3 Hz), 3.46 (s, 3H, OCH_3), 3.66 (d, 2H, $J_{5,4}$=3.7 Hz, H-5a, H-5b), 4.18 (apparent q, 1H, H-4), 4.52 (s, 1H, H-1), 4.60 (s, 2H, CH_2Ph), 4.63, 4.81 (2d, 2H, J_{gem}=13.2 Hz, CH_2Ph), 7.19–7.26, 7.34–7.43 (2m, 10H, 2Ph).

^{13}C NMR ($CDCl_3$): δ 24.88, 55.45, 69.95, 70.24, 70.88, 77.06, 82.18, 83.01, 107.63, 127.32, 129.36, 130.01, 130.32, 133.68, 133.78, 134.13, 134.18, 134.45, 134.58.

Step D: 4-Chloro-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C-methyl-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine

To a solution of the compound from Step C (9.42 g, 19 mmol) in anhydrous dichloromethane (285 mL) at 0° C. was added HBr (5.7 M in acetic acid, 20 mL, 114 mmol) dropwise. The resulting solution was stirred at 0° C. for 1 h and then at room temperature for 3 h, evaporated in vacuo and co-evaporated with anhydrous toluene (3×40 mL). The oily residue was dissolved in anhydrous acetonitrile (50 mL) and added to a solution of sodium salt of 4-chloro-1H-pyrrolo[2,3-d]pyrimidine [for preparation, see *J. Chem. Soc.*, 131 (1960)] in acetonitrile [generated in situ from 4-chloro-1H-pyrrolo[2,3-d]pyrimidine (8.76 g, 57 mmol) in anhydrous acetonitrile (1000 mL), and NaH (60% in mineral oil, 2.28 g, 57 mmol), after 4 h of vigorous stirring at room temperature]. The combined mixture was stirred at room temperature for 24 h, and then evaporated to dryness. The residue was suspended in water (250 mL) and extracted with EtOAc (2×500 mL). The combined extracts were washed with brine (300 mL), dried over Na_2SO_4, filtered and evaporated. The crude product was purified on a silica gel column (10 cm×10 cm) using ethyl acetate/hexane (1:3 and 1:2) as the eluent. Fractions containing the product were combined and evaporated in vacuo to give the desired product (5.05 g) as a colorless foam.

^{1}H NMR ($CDCl_3$): δ 0.93 (s, 3H, CH_3), 3.09 (s, 1H, OH), 3.78 (dd, 1H, $J_{5',5''}$=10.9 Hz, $J_{5',4}$=2.5 Hz, H-5'), 3.99 (dd, 1H, $J_{5'',4}$=2.2 Hz, H-5"), 4.23–4.34 (m, 2H, H-3', H-4'), 4.63, 4.70 (2d, 2H, J_{gem}=12.7 Hz, CH_2Ph), 4.71, 4.80 (2d, 2H, J_{gem}=12.1 Hz, CH_2Ph), 6.54 (d, 1H, , $J_{5,6}$=3.8 Hz, H-5), 7.23–7.44 (m, 10H, 2Ph).

^{13}C NMR ($CDCl_3$): δ 21.31, 69.10, 70.41, 70.77, 79.56, 80.41, 81.05, 91.11, 100.57, 118.21, 127.04, 127.46, 127.57, 129.73, 129.77, 130.57, 130.99, 133.51, 133.99, 134.33, 134.38, 134.74, 135.21, 151.07, 151.15 152.47.

Step E: 4-Chloro-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

To a solution of the compound from Step D (5.42 g, 8.8 mmol) in dichloromethane (175 mL) at −78° C. was added boron trichloride (1M in dichloromethane, 88 mL, 88 mmol) dropwise. The mixture was stirred at −78° C. for 2.5 h, then at −30° C. to −20° C. for 3 h. The reaction was quenched by addition of methanol/dichloromethane (1:1) (90 mL) and the resulting mixture stirred at −15° C. for 30 min., then neutralized with aqueous ammonia at 0° C. and stirred at room temperature for 15 min. The solid was filtered and washed with CH_2Cl_2/MeOH (1/1, 250 mL). The combined filtrate was evaporated, and the residue was purified by flash chromatography over silica gel using CH_2Cl_2 and CH_2Cl_2:MeOH (99:1, 98:2, 95:5 and 90:10) gradient as the eluent to furnish desired compound (1.73 g) as a colorless foam, which turned into an amorphous solid after treatment with MeCN.

^{1}H NMR (DMSO-d_6) δ 0.64 (s, 3H, CH_3), 3.61–3.71 (m, 1H, H-5'), 3.79–3.88 (m, 1H, H-5"), 3.89–4.01 (m, 2H, H-3', H-4'), 5.15–5.23 (m, 3H, 2'-OH, 3'-OH, 5'-OH), 6.24 (s, 1H, H-1'), 6.72 (d, 1H, $J_{5,6}$=3.8 Hz, H-5), 8.13 (d, 1H, H-6), 8.65 (s, 1H, H-2).

^{13}C NMR (DMSO-d_6) δ 20.20, 59.95, 72.29, 79.37, 83.16, 91.53, 100.17, 117.63, 128.86, 151.13, 151.19, 151.45.

Step F: 4-Amino-7-(2-C-methyl-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

To the compound from Step E (1.54 g, 5.1 mmol) was added methanolic ammonia (saturated at 0° C.; 150 mL). The mixture was heated in a stainless steel autoclave at 85° C. for 14 h, then cooled and evaporated in vacuo. The crude mixture was purified on a silica gel column with CH_2Cl_2/MeOH (9/1) as eluent to give the title compound as a colorless foam (0.8 g), which separated as an amorphous solid after treatment with MeCN. The amorphous solid was recrystallized from methanol/acetonitrile; m.p. 222° C.

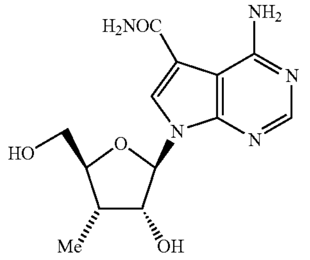
^{1}H NMR (DMSO-d_6): δ 0.62 (s, 3H, CH_3), 3.57–3.67 (m, 1H, H-5'), 3.75–3.97 (m, 3H, H-5", H-4', H-3'), 5.00 (s, 1H, 2'-OH), 5.04 (d, 1H, $J_{3'OH,3'}$=6.8 Hz, 3'-OH), 5.06 (t, 1H, $J_{5'OH,5',5''}$=5.1 Hz, 5'-OH), 6.11 (s, 1H, H-1'), 6.54 (d, 1H, $J_{5,6}$=3.6 Hz, H-5), 6.97 (or s, 2H, NH_2), 7.44 (d, 1H, H-6), 8.02 (s, 1H, H-2).

^{13}C NMR (DMSO-d_6): δ 20.26, 60.42, 72.72, 79.30, 82.75, 91.20, 100.13, 103.08, 121.96, 150.37, 152.33, 158.15.

LC-MS: Found: 279.10 (M−H^+); calc. for $C_{12}H_{16}N_4O_4$+H^+: 279.11.

EXAMPLE 63

4-Amino-7-(3-deoxy-3-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide



Step A: 4-Amino-6-bromo-7-(2-O-acetyl-5-O-benzoyl-3-deoxy-3-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile

BSA (0.29 mL, 2.0 mmol) was added into a stirred suspension of 4-amino-6-bromo-5-cyano-1H-pyrrolo[2,3-d]pyrimidine (0.24 g, 1 mmol; prepared according to *Nucleic Acid Chemistry*, Part IV, Townsend, L. B. and Tipson, R. S.; Ed.; Wiley-Interscience: New York, 1991, pp. 16–17 and *Synthetic Commun*. 1998, 28, 3835) in dry acetonitrile (10 mL) at room temperature under argon. After 15 min, 1,2-di-O-acetyl-5-O-benzoyl-3-deoxy-3-methyl-D-ribofuranose (J. Med. Chem. (1976), 19, 1265) (0.36 g, 1.0 mmol) was added along with TMSOTf (0.54 g, 3 mmol). The mixture was stirred at room temperature for 5 min and then at 80° C. for 0.5 h. The solution was cooled, diluted with ethyl acetate (50 mL) and poured into ice-cold saturated aqueous $NaHCO_3$ (15 mL). The layers were separated. The organic layer was washed with brine (15 mL), dried (Na_2SO_4) and then evaporated. The residue was purified on silica gel column using a solvent system of hexanes/EtOAc: 3/1. Appropriate fractions were collected and evaporated to provide the title compound as colorless foam (0.21 g).

Step B: 4-Amino-7-(2-O-acetyl-5-O-benzoyl-3-deoxy-3-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile

To a suspension of the title compound from Step A (183 mg, 0.35 mmol) in EtOH (9 mL) were added ammonium formate (0.23 g, 3.6 mmol) and 10% palladium on activated carbon (20 mg) and the mixture was heated at reflux for 1.5 h. The hot reaction mixture was filtered through Celite and washed with hot EtOH. The solvent was removed and the residue treated with MeOH. The pale yellow solid was filtered thus yielding 105 mg of pure title compound. The filtrate was evaporated and purified on a silica gel column with a solvent system of CH_2Cl_2/MeOH: 50/1 to afford an additional 63 mg of title compound as a white solid.

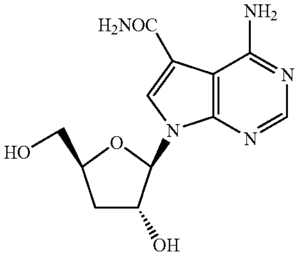
Step C: 4-Amino-7-(3-deoxy-3-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide

A mixture of the compound from Step B (51 mg, 0.12 mmol), ethanolic ammonia (5 mL, saturated at 0° C.), aqueous ammonia (5 mL, 30%) and aqueous hydrogen peroxide (1 mL, 35%) was stirred room temperature for 8 h. The solution was evaporated and:the residue purified on silica gel column with a solvent system of CH_2Cl_2/MeOH: 10/1 to give the title compound as a white solid (28 mg).

^{1}H-MNR (CD_3OD): δ 1.12 (d, 3H, J=6.8 Hz), 2.40 (m, 1H), 3.76 (dd, 1H, J_1=12.8 Hz, J_2=4.0 Hz), 3.94–4.04 (m, 2H), 4.33 (d, 1H, J=5.4 Hz), 6.13 (s, 1H), 8.11 (s, 1H), 8.16 (s, 1H).

EXAMPLE 64

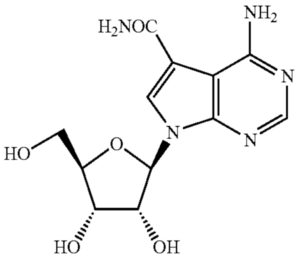
4-Amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carboxamide



This compound was prepared following the procedures described in *J. Med. Chem.* 26: 25 (1983).

EXAMPLE 65

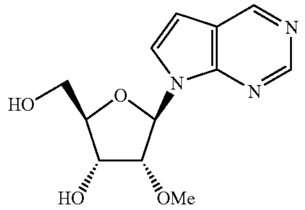
4-Amino-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carboxamide (Sangivamycin)



This compound was obtained from commercial sources.

EXAMPLE 66

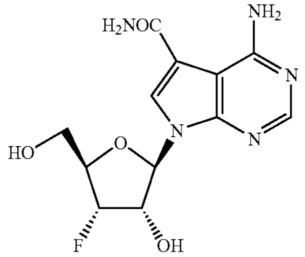
7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



This compound was prepared following the procedures described in *J. Org. Chem.* 39: 1891 (1974).

EXAMPLE 67

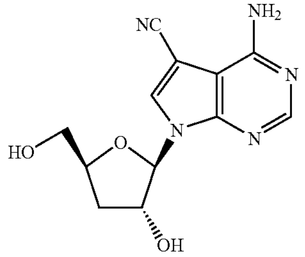
4-Amino-7-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carboxamide



This compound was prepared following the procedures described in *Chem. Pharm. Bull.* 41: 775 (1993).

EXAMPLE 68

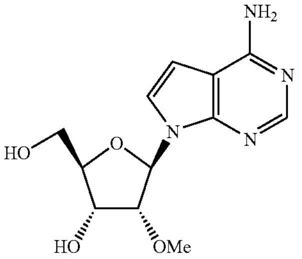
4-Amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile



This compound was prepared following the procedures described in *J. Med. Chem.* 30: 481 (1987).

EXAMPLE 69

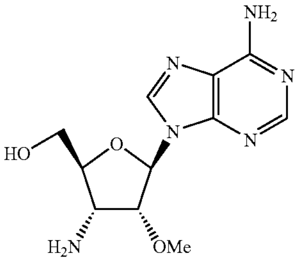
4-Amino-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



This compound was prepared following the procedures described in *J. Org. Chem.* 39: 1891 (1974).

EXAMPLE 70

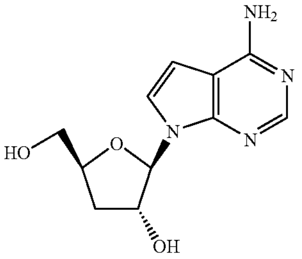
3'-Amino-3'-deoxy-2'-O-methyladenosine



This compound is obtained by the methylation of appropriately protected 3'-amino-3'-deoxyadenosine derivative (Example 54).

EXAMPLE 71

4-Amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



This compound was prepared following the following procedure described in *Can. J. Chem.* 55: 1251 (1977).

EXAMPLE 72

General Process to SATE Prodrug Moiety

S-Acyl-2-Thioethyl (SATE) pronucleotides are discussed in C. R. Wagner, V. V. Iyer, and E. J. McIntee, "Pronucleotides: Toward the In Vivo Delivery of Antiviral and Anticancer Nucleotides," *Med. Res. Rev.*, 20: 1–35 (2000), which is incorporated by reference herein in its entirety. SATE derivatives of nucleosides are also disclosed U.S. Pat. Nos. 5,770,725; 5,849,905; and 6,020,482, the contents of each of which are incorporated by reference herein in their entirety.

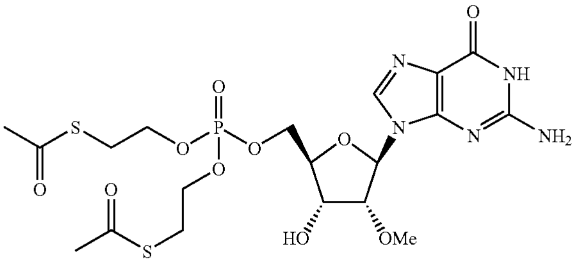
Bis(S-acetyl-2-thioethyl)-N,N-diisopropylphosphoramidite:

2-Mercaptoethanol (5 g, 64 mmol) was dissolved in CH_2Cl_2 (50 mL). To this solution was added triethylamine (7.67 mL, 57.6 mmol), and the reaction mixture was cooled in an ice bath to 0° C. Acetic anhydride (4.54 mL, 48 mmol) was added dropwise in 10 min, and the reaction mixture was stirred for 1 h at 0° C. The reaction mixture was then allowed to come to room temperature over a period of 2 h. The reaction mixture was diluted with CH_2Cl_2 (50 mL), washed with water (75 mL), 5% aqueous $NaHCO_3$ (75 mL) and brine (75 mL). The organic phase was dried over anhydrous Na_2SO_4 and concentrated in vacuo to give an oil. The oil was then dissolved in anhydrous THF (40 mL) and anhydrous triethylamine (7.76 mL) was, added. To this mixture was added activated molecular sieves (4Å) and was kept at room temperature for 10 min. The reaction mixture was cooled in an ice bath to 0° C. and diisopropylphosphoramidous dichloride (6.47 g, 32.03 mmol) was added. The reaction mixture was stirred at 0° C. for 2 h under inert atmosphere. Hexane (40 mL) was added to the reaction mixture and the precipitate formed was filtered. The filtrate was concentrated to one fourth of the volume, purified by loaded silica gel column chromatography and eluted with hexane containing 3% triethylamine and incremental amount of ethyl acetate (0 to 7%) to give the title compound as an oil (2.36 g).

^1H NMR ($CDCl_3$): δ 1.17 (s, 6H), 1.21 (s, 6H), 2.36 (s, 6H), 3.14 (t, J=6.44 Hz), 3.51–3.84 (m, 6H); ^{13}C NMR ($CDCl_3$): δ 24.47, 24.61, 30.48, 42.85, 43.1, 61.88, 62.23, 195.26; ^{13}P NMR ($CDCl_3$): δ 146.96.

EXAMPLE 73

2'-O-Methylguanosine-5'-[bis-(S-acetyl-2-thioethyl) phosphate]



Step A: N^{2}-(4-monomethoxytrityl)-2'-O-methylguanosine-5'-[bis-(S-acetyl-2-thioethyl)phosphate]

N^{2}-(4-monomethoxytrityl)-2'-O-methylguanosine (0.74 g, 1.31 mmol) was mixed with 1H-tetrazole (0.061 g, 0.87 mmol) and dried over P_2O_5 in vacuo overnight. To this mixture was added anhydrous acetonitrile (8 mL). To the turbid solution, bis(S-acetyl-2-thioethyl)N,N-diisopropylphosphoramidite (0.3 g, 0.87 mmol) was added slowly and the reaction mixture was stirred at ambient temperature under inert atmosphere for 2 h. Solvent was removed in vacuo. The residue was cooled to −40° C. and a solution of 3-chloroperbenzoic acid (0.2 g) in CH_2Cl_2 (7 mL) was added. The solution was allowed to warm up to room temperature over 1 h. Sodium hydrogensulfite (10% aqueous solution, 2 mL) was added to reduce the excess of 3-chloroperbenzoic acid. The organic phase separated, diluted with CH_2Cl_2 (20 mL), washed with saturated aqueous Na_2CO_3 (10 mL), water (10 mL), dried over Na_2SO_4 and evaporated to dryness. The residue was purified by silica gel column chromatography and eluted with CH_2Cl_2 containing incremental amount of MeOH (5 to 10%) as eluent to yield the title compound (0.36 g) as a foam.

^{1}H NMR (MSO-d$_6$): δ 2.35 (s, 6H), 2.97 (s, 3H), 3.11 (t, 4H, J=6.0 Hz), 3.5 (m, 1H), 3.74 (s, 3H), 3.72–3.83 (m, 2H), 3.97–4.11 (m, 6H), 5.1 (d, 1H, J=6.4 Hz), 5.29 (d, 1H, J=3.1 Hz), 6.89 (d, 2H, J=8.8 Hz), 7.15–7.37 (m, 12H), 7.68 (s, 1H), 7.73 (s, 1H), 10.72 (s, 1H); ^{13}C NMR (CDCl$_3$): δ 30.36, 55.38, 57.99, 66.08, 66.19, 67.22, 69.15, 70.49, 81.18, 81.57, 86.64, 113.04, 117.99, 126.66, 127.71, 128.67, 130.04, 136.09, 136.56, 144.51, 144.82, 149.52, 151.29, 158.15, 194.56; ^{13}P NMR (CDCl$_3$): δ −2.04; MS (API-ES) 852.10 [M−H]$^+$.

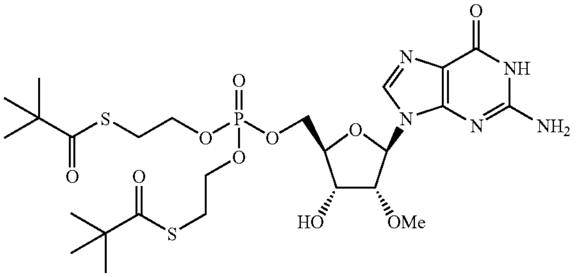
Step B: 2'-O-methylguanosine-5'-[bis-(S-acetyl-2-thioethyl) phosphate]

N^{2-}(4-monomethoxytrityl)-2'-O-methylguanosine-5'-[bis-(S-acetyl-2-thioethyl)phosphate] (0.2 g, 0.23 mmol) was dissolved in acetic acid: MeOH:H$_2$O, 3:6:1 and heated at 55° C. for 24 h. Solvent was removed and the residue was purified by HPLC on reverse phase column (Hamilton PRP-1, 250×22 mm, A=Acetonitrile, B=H$_2$O 20 to 100 B in 65 min, flow 10 mL min^{-1}). Fractions containing the product were pooled together and evaporated to give the title compound (40% yield).

^{13}P NMR (CDCl$_3$): δ −0.72; MS (API-ES) m/z 582.1 [M+H]$^+$.

EXAMPLE 74

2'-O-Methylguanosine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate]



Step A: Bis(S-pivaloyl-2-thioethyl)-N,N-diisopropylphosphoramidite

S-pivaloyl-2-thioethanol (6.3 g, 39.6 mmol) was dissolved in anhydrous THF (100 mL). To this solution was added activated molecular sieves (4 Å) and kept at room temperature for 30 min. Anhydrous triethylamine (7.9 mL, 59.4 mmol) was added and the reaction mixture was cooled in an ice bath to 0° C. To this mixture diisopropylphosphoramidous dichloride (4 g, 19.8 mmol) was added dropwise. The mixture was stirred the reaction mixture at 0° C. for 2 h under inert gas atmosphere. Hexane (100 mL) was added to the reaction mixture, and the precipitate formed was filtered. The filtrate was concentrated to one fourth of the volume. This was purified by flash silica gel column chromatography using hexane containing 2% triethylamine and incremental amount of ethyl acetate (0 to 3%) as eluent to give the title compound as an oil (5.23 g).

^{1}H NMR (CDCl$_3$): δ 1.13–1.31 (m, 30H), 1.21 (s, 6H), 3.09 (t, J=6.6 Hz, 4H), 3.51–3.84 (m, 6H); ^{13}C NMR (CDCl$_3$): δ 24.47, 24.61, 27.32, 30.00, 42.85,43.1, 46.32, 61.98, 62.33, 206.1; ^{13}P NMR (CDCl$_3$): δ 148.51.

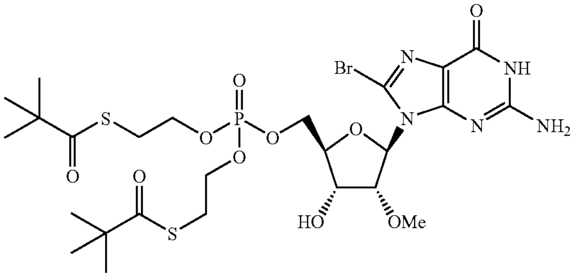
Step B: 2'-O-methylguanosine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate]

N^2-(4-monomethoxytrityl)-2'-O-methylguanosine (0.6 g, 1.05 mmol) was mixed with 1H-tetrazole (0.05 g, 0.7 mmol) and dried over P$_2$O$_5$ in vacuo overnight. To this mixture anhydrous acetonitrile (13.8 mL) was added. The reaction mixture was cooled to 0° C. in an ice bath and bis(S-pivaloyl-2-thioethyl)N,N-diisopropylphosphoramidite (0.32 g, 0.7 mmol) was added slowly. The reaction mixture was stirred at 0° C. for 5 minutes. The ice bath was removed and the reaction mixture was allowed to stir at room temperature under an inert atmosphere for 2 h. Solvent was removed in vacuo. The residue was cooled to 40° C. and a solution of 3-chloroperbenzoic acid (0.24 g, 1.4 mmol, 57–80%) in CH$_2$Cl$_2$ (10 mL) was added. The solution was allowed to warm up to −10° C. over 1 h. Sodium hydrogensulfite (10% aqueous solution, 10 mL) was added to reduce the excess of 3-chloroperbenzoic acid. The organic phase separated, diluted with CH$_2$Cl$_2$ (50 mL), washed with saturated aqueous Na$_2$CO$_3$ (40 mL), water (40 mL), dried over Na$_2$SO$_4$ and evaporated to dryness. The residue was chromatographed on a flash silica gel column using a CH$_2$Cl$_2$ containing incremental amount of MeOH (0 to 5%) as eluent. Fractions containing the product were pooled together and evaporated. The residue was dissolved in a solution of acetic acid/water/methanol (10 mL, 3:1:6) and heated at 55° C. for 24 h. Evaporated the solution in vacuum to get an oil. The oil was dissolved in 20% MeOH in water and purified by HPLC on C-18 column (Luna C-18, 250×2.12 mm, A=water, B=acetonitrile, 20 to 10% B in 65 min., flow 10 mL min^{-1}, λ 260 nm) to yield the title compound (0.082 g).

^{1}H NMR (DMSO-d$_6$): δ 1.18 (s, 18H), 3.08 (m, 4H), 3.33 (s, 3H) 3.94–4.10 (m, 6H), 4.14–4.21 (m, 2H), 4.29 (m, 1H), 5.42 (d, 1H, J=5.4 Hz), 5.81 (d, 1H, J=5.8 Hz), 6.49 (bs, 2H), 7.86 (s, 1H), 10.66 (bs, 1H); ^{13}P NMR (DMSO-d$_6$): δ −0.71; MS (API-ES) m/z 664.2 [M−H]$^-$.

EXAMPLE 75

8-Bromo-2'-O-methylguanosine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate]

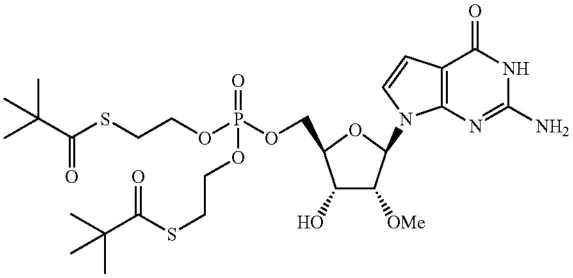


This compound was synthesized according to the procedure used for the synthesis of Example 74 starting with 8-bromo-N^2-(4-monomethoxytrityl)-2'-O-methylguanosine (0.46 g, 0.63 mmol). Other reagents used were 1H-tetrazole (0.034 g, 0.49 mmol), bis(S-pivaloyl-2-thioethyl)N,N-diisopropylphosphoramidite (0.22 g, 0.49 mmol), acetonitrile (8.3 ml), 3-chloroperbenzoic acid (0.17 g, 0.98 mmol, 57–80%) in CH$_2$Cl$_2$ (4 mL). The title compound was isolated in 13% yield (0.061 g).

^{1}H NMR (DMSO-d$_6$): δ 1.14 and 1.16 (m, 18H), 3.06 (m, 4H), 3.32 (s, 3H) 3.96–4.06 (m, 5H), 4.18–4.3 (m, 2H), 4.46 (d, 1H, J=2.4 Hz), 4.66 (t, 1H, J=2.6 Hz), 5.37 (d, 1H, J=2.6 Hz), 5.78 (d, 1H, J=2.8 Hz), 6.62 (bs, 2H), 10.99 (bs, 1H); ^{13}P NMR (DMSO-d$_6$) δ −0.79; MS (API-ES) m/z 742.13 and 744.13 [M−H]$^-$.

EXAMPLE 76

2-Amino-3,4-dihydro-7-(2-O-methyl-β-D-ribofuranosyl)-4-oxo-7H-pyrrolo[2,3-d]pyrimidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate]

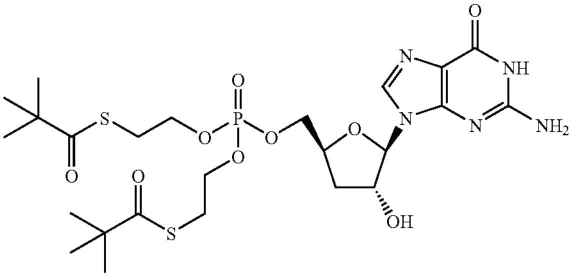


This compound was synthesized according to the procedure used for the synthesis of Example 74 starting with 7-deaza-N^2-(4-monomethoxytrityl)-2'-O-methylguanosine (0.47 g, 0.82 mmol). Other reagents used were 1H-tetrazole (0.044 g, 0.63 mmol), bis(S-pivaloyl-2-thioethyl)N,N-diisopropylphosphoramidite (0.29 g, 0.63 mmol), acetonitrile (11 mL), 3-chloroperbenzoic acid (0.21 g, 1.26 mmol, 57–80%) in CH_2Cl_2 (5.2 mL). The title compound was isolated in 29% yield (0.158 g).

^{1}H NMR (DMSO-d$_6$): δ 1.14 (s, 18H), 3.06 (m, 4H), 3.31 (s, 3H) 3.96–4.26 (m, 9H), 5.35 (d, 1H, J=2.6 Hz), 5.78 (d, 1H, J=5.2 Hz), 5.99 (d, 1H, J=6.6 Hz), 6.27 (m, 3H), 6.86 (d, 1H, J=3.6 Hz), 10.39 (s, 1H); ^{13}P NMR (DMSO-d$_6$): δ −0.72; MS (API-ES) m/z 663.20 [M−H]$^-$; HRMS Calcd for $C_{26}H_{42}N_4O_{10}PS_2$ 665.2074 found 665.2071.

EXAMPLE 77

3'-Deoxyguanosine-5'-[bis-(S-pivaloyl-2-thioethyl) phosphate]

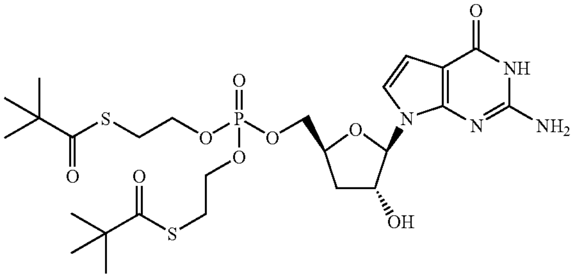


N^2-(4-Monomethoxytrityl)-3'-deoxyguanosine (0.20 g, 0.35 mmol) was mixed with 1H-tetrazole (0.019 g, 0.27 mmol) and dried over P_2O_5 in vacuo overnight. To this mixture anhydrous acetonitrile (4.7 mL) was added to give a turbid solution. The reaction mixture was cooled to 0° C. in an ice bath and bis(S-pivaloyl-2-thioethyl)N,N-diisopropylphosphoramidite (0.12 g, 0.27 mmol) was added slowly. The reaction mixture was stirred at 0° C. for 5 minutes. The ice bath was removed and the reaction mixture was allowed to come to room temperature. The reaction mixture was stirred at room temperature under an inert gas atmosphere for 2 h. Solvent was removed in vacuo. The residue was cooled to 40° C. and a solution of 3-chloroperbenzoic acid (0.12 g, 0.7 mmol, 57–80%) in CH_2Cl_2 (2.2 mL) was added. The solution was allowed to warm up to −10° C. over 1 h. Sodium hydrogensulfite (10% aqueous solution, 2 mL) was added to reduce the excess of 3-chloroperbenzoic acid. The organic phase was separated, diluted with CH_2Cl_2 (30 mL), washed with saturated aqueous Na_2CO_3 (20 mL), water (20 mL), dried over Na_2SO_4 and evaporated to dryness. The residue was chromatographed on a flash silica gel column using CH_2Cl_2 containing incremental amount of MeOH (0 to 5%) as eluent. Fractions containing the product were pooled and evaporated. The residue was dissolved in a solution of acetic acid/water/methanol (5 mL, 3:1:6) and heated at 55° C. for 24 h. Evaporated the solution in vacuum to get an oil. The oil was dissolved in 20% MeOH in water and purified by HPLC on C-18 column (Luna C-18, 250×2.12 mm, A=water, B=acetonitrile, 20 to 10% B in 65 min., flow 10 mL min^{-1}, λ 260 nm) to yield the title compound (0.027 g).

^{1}H NMR (DMSO-d$_6$): δ 1.15 (s, 18H), 1.92–2.01 (m, 1H), 2.17–2.28 (m, 1H), 3.04 (t, 4H, J=6.2 Hz), 3.91–4.23 (m, 6 H), 4.37–4.55 (m, 2H), 5.67 (m, 2H), 6.45 (bs, 2H), 7.75 (s, 1H), 10.61 (s, 1H); ^{13}P NMR (DMSO-d$_6$): δ −0.75; MS (API-ES) m/z 634.2 [M−H]$^-$.

EXAMPLE 78

2-Amino-7-(3-deoxy-β-D-ribofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo[2,3-d]pyrimidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate]

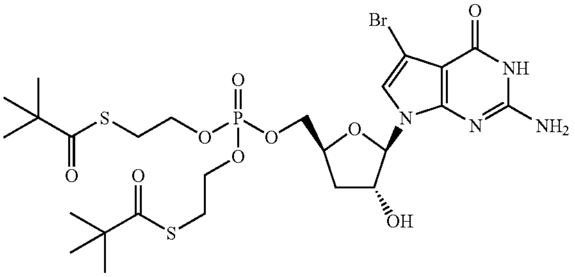


2-(4-Monomethoxytrityl)amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one (0.30 g, 0.52 mmol) was mixed with 1H-tetrazole (0.028 g, 0.40 mmol) and dried over P_2O_5 in vacuo overnight. To this mixture anhydrous acetonitrile (7 mL) was added, and the solution was cooled to 0° C. in an ice bath. Bis(S-pivaloyl-2-thioethyl)-N,N-diisopropylphosphoramidite (0.18 g, 0.40 mmol) was added slowly. The reaction mixture was allowed to come to at room temperature and stirred at room temperature under an inert atmosphere for 2 h. The solvent was removed in vacuo. The residue was cooled to 40° C., and a solution of 3-chloroperbenzoic acid (0.14 g, 0.8 mmol, 57–80%) in CH_2Cl_2 (5 mL) was added. The solution was allowed to warm up to −10° C. over 2 h. Sodium hydrogensulfite (10% aqueous solution, 5 mL) was added to reduce the excess of 3-chloroperbenzoic acid. The organic phase was separated, diluted with CH_2Cl_2 (50 mL), washed with saturated aqueous Na_2CO_3 (40 mL), water (40 mL), dried over Na_2SO_4 and evaporated to dryness. The residue was chromatographed on a flash silica gel column using CH_2Cl_2 containing incremental amount of MeOH (0 to 5%) as eluent. Fractions containing the product were pooled and evaporated. The residue was dissolved in a solution of acetic acid/water/methanol (10 mL, 3:1:6) and heated at 55° C. for 24 h. The solution was evaporated to give an oil. The oil was dissolved in 20% MeOH in water and purified by HPLC on C-18 column (Luna C-18,250×2.12 mm, A=water, B=acetonitrile 20 to 10% B in 65 mL, flow 10 mL/min, λ 260 nm) to give the title compound (0.053 g).

^{1}H NMR (DMSO-d$_6$): δ 1.16 (s, 18H), 1.91–2.01 (m, 1H), 2.17–2.25 (m, 1H), 3.05 (t, 4H, J=6.2 Hz), 3.92–4.2 (m, 6H), 4.35 (bs, 2H), 5.56 (d, 1H, J=4.2 Hz), 5.86 (d, 1H, J=2.4 Hz), 6.24 (m, 3H), 6.77 (d, 1H, J=3.6 Hz), 10.36 (s, 1H); ^{13}P NMR DMSO-d$_6$): δ −0.89; HRMS (MALDI) Calcd for $C_{25}H_{39}N_4O_9PS_2$.635.1969 found 635.1964.

EXAMPLE 79

2-Amino-5-bromo-7-(3-deoxy-β-D-ribofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo[2,3-d]pyrimidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate]



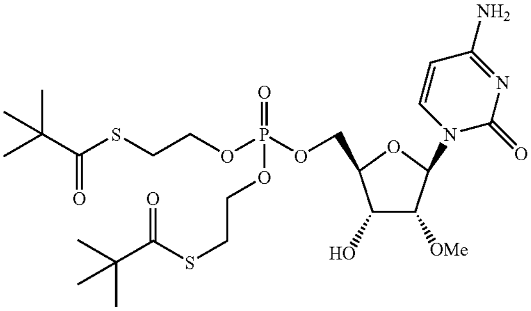
2-(4-Monomethoxytrityl)amino-5-bromo-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one (0.066 g, 0.17 mmol) was mixed with imidazole triflate (0.017 g, 0.17 mmol) and dried over P_2O_5 in vacuo overnight. To this mixture anhydrous acetonitrile (7 mL) and bis(S-pivaloyl-2-thioethyl)N,N-diisopropyl- phosphoramidite (0.97 g, 0.24 mmol) were added slowly. The reaction mixture was stirred under an inert atmosphere for 18 h. Solvent was removed in vacuo. The residue was cooled to −40° C. and a solution of 3-chloroperbenzoic acid (0.059 g, 0.34 mmol, 57–80%) in CH_2Cl_2 (2 mL) was added. The solution was allowed to warm up to −10° C. over 2 h. Sodium hydrogensulfite (10% aqueous solution, 5 mL) was added to reduce the excess of 3-chloroperbenzoic acid. The organic phase was separated, diluted with CH_2Cl_2 (30 mL), washed with saturated aqueous Na_2CO_3 (20 mL), water (20 mL), dried over Na_2SO_4 and evaporated to dryness. The residue was chromatographed on flash silica gel column using CH_2Cl_2 containing incremental amount of MeOH (0 to 5%) as eluent. Fractions containing the product were pooled and evaporated. The residue was dissolved in a solution of acetic acid/water/methanol (3 mL, 3:1:6) and heated at 55° C. for 24 h. The solution was evaporated to give an oil. The oil was dissolved in 20% MeOH in water and purified by HPLC on C-18 column (Luna C-18,250×2.12 mm, A=water, B=acetonitrile 20 to 10% B in 65 mL, flow 10 mL min^{-1}, λ 260 nm) to afford the title compound (0.036 g).

^{1}H NMR (DMSO-d$_6$): δ 1.17 (s, 18H), 1.87–2.03 (m, 1H), 2.17–2.26 (m, 1H), 3.05 (t, 4H, J=6.4 Hz), 3.92–4.2 (m, 6H), 4.37 (bs, 2H), 5.70 (d, 1H, J=4.4 Hz), 5.85 (d, 1H, J=2.6 Hz),

6.36 (bs, 2H), 6.93 (s, 1H), 10.51 (s, 1H); ^{13}P NMR (DMSO-d$_6$): δ −0.89; MS (AP-ES) m/z 711.11 and 713.09 $[M-H]^-$; HRMS (MALDI) Calcd for $C_{25}H_{38}BrN_4O_9PS_2$.713.1074 and 715.1074 found 713.1081 and 715.102.

EXAMPLE 80

2'-O-Methylcytidine-5'-[bis-(S-pivaloyl-2-thioethyl) phosphate]

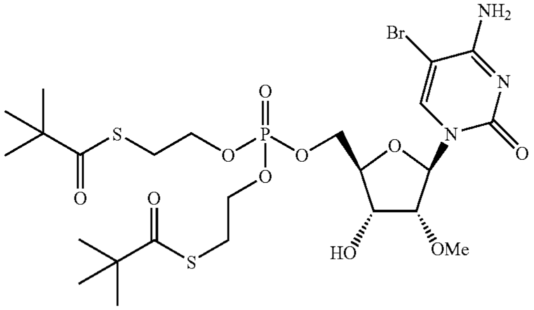


N^4-(4,4'-Dimethoxytrityl)-2'-O-methylcytidine (0.49 g, 0.86 mmol) was mixed with 1H-tetrazole (0.06 g, 0.86 mmol) and dried over P_2O_5 in vacuo overnight. To this mixture anhydrous acetonitrile (6 mL) and bis-(S-pivaloyl-2-thioethyl)-N,N-diisopropylphosphoramidite (0.39 g, 0.86 mmol) were added at 0° C. The reaction mixture was allowed to come to room temperature and stirred under an inert atmosphere for 18 h. Solvent was removed in vacuo. The residue was cooled to −40° C. and a solution of 3-chloroperbenzoic acid (0.3 g, 1.72 mmol, 57–80%) in CH_2Cl_2 (5.5 mL) was added. The solution was allowed to warm up to −10° C. over 2 h. Sodium hydrogensulfite (10% aqueous solution, 5 mL) was added to reduce the excess of 3-chloroperbenzoic acid. The organic phase was separated, diluted with CH_2Cl_2 (30 mL), washed with saturated aqueous Na_2CO_3 (20 mL), water (20 mL), dried over Na_2SO_4 and evaporated to dryness. The residue was chromatographed on a flash silica gel column using CH_2Cl_2 containing incremental amount of MeOH (0 to 10%) as eluent. Fractions containing the product were pooled and evaporated. The residue was dissolved in a solution of acetic acid/water/methanol (10 mL, 3:1:6) and heated at 55° C. for 24 h. The solution was evaporated to give an oil. The oil was dissolved in 20% MeOH in water and purified by HPLC on C-18 column (Luna C18,250×2.12 mm, A=water, B=acetonitrile 20 to 10% B in 65 mL, flow 10 mL min^{-1}, λ 260 nm) to yield the title compound (0.076 g).

^{1}H NMR (DMSO-d$_6$): δ 1.18 (s, 18H), 3.12 (t, 4H, J=6.4 Hz), 3.39 (s, 3H), 3.69 (t, 1H, J=4.2 Hz), 3.93–4.3 (m, 8H), 5.29 (d, 1H, J=6.2 Hz), 5.72 (d, 1H, J=7.4 Hz), 5.86 (d, 1H, J=4 Hz), 7.21 (bs, 2H), 7.58 (d, 1H, J=7.4 Hz); ^{13}P NMR (CD$_3$CN): δ −0.64; MS (AP-ES) m/z 625.69 $[M+H]^+$; HRMS (MALDI) Calcd for $C_{24}H_{40}N_3O_{10}PS_2Na$ 648.1785 found 648.1804.

EXAMPLE 81

5-Bromo-2'-O-methylcytidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate]



Step A: 5-Bromo-3'-O-(t-butyldimethyl)silyl-2'-O-methylcytidine

2'-O-Methylcytidine (1.5 g, 5.83 mmol) was mixed with imidazole (3.97 g, 58.32 mmol) and dried in vacuo. This mixture was dissolved in anhydrous DMF (4 mL) and t-butyldimethylsilyl chloride (4.41 g, 29.25 mmol) was added and the reaction mixture was stirred for 18 h at room temperature under an inert atmosphere. Reaction mixture was diluted with water (100 mL) and extracted with ethyl acetate (2×60 mL). The organic phase was dried over anhydrous Na_2SO_4 and evaporated. The residue was purified by silica gel column chromatography and eluted with ethyl acetate/hexane, 6:4. Fractions containing the product were pooled and evaporated. The product obtained (2.76 g) was dissolved in acetonitrile (19.43 mL), LiBr (0.623 g, 7.18 mmol) and stirred to get a clear solution. To this ammonium ceric (IV) nitrate (6.24 g, 11.37 mmol) was added and the reaction mixture was allowed to stir at room temperature for 3 h. Solvent was removed in vacuum. The residue obtained was taken in ethyl acetate (100 mL) and washed with water (80 mL). The organic phase was separated, dried over anhydrous Na_2SO_4 and evaporated. Residue purified by silca gel column chromatography and eluted with 5% MeOH in CH_2Cl_2. The product obtained (2.66 g) was dissolved in 80% acetic acid in water and heated at 50° C. for 6 h. The solvent was removed and the residue purified on a silica gel column and eluted with 5% MeOH in CH_2Cl_2 to give the title compound (0.85 g).

^1H NMR (DMSO-d_6): δ 0.78 (s, 6H), 0.85 (s, 9H), 3.31 (s, 3H), 3.44–3.6 (m, 2H), 3.69–3.9 (m, 2H), 4.24 (m, 1H), 5.29 (t, 1H, J=4.4 Hz), 5.76 (d, 1H, J=3.2 Hz), 7.06 (bs, 1H), 7.88 (bs, 1H), 8.39 (s, 1H).

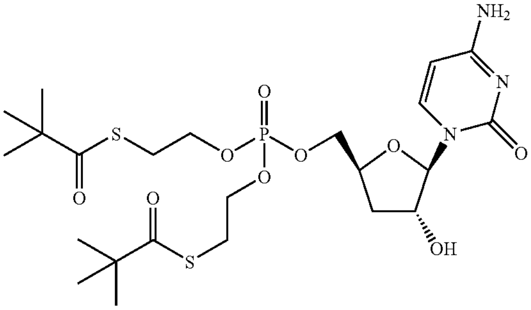
Step B: 5-Bromo-2'-O-methylcytidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate]

5-Bromo-3'-O-(t-butyldimethyl)silyl-2'-O-methylcytidine (0.093 g, 0.21 mmol) was mixed with 1H-tetrazole (0.03 g, 0.42 mmol) and dried over P_2O_5 in vacuo overnight. To this mixture anhydrous acetonitrile (2 mL). Bis-(S-pivaloyl-2-thioethyl)-N,N-diisopropylphosphoramidite (0.2 g, 0.42 mmol) was added at 0° C. The reaction mixture was allowed to come to room temperature and stirred under an inert atmosphere for 4 h. Solvent was removed in vacuo. The residue was cooled to −40° C. and a solution of 3-chloroperbenzoic acid (0.072 g, 0.42 mmol, 57–80%) in CH_2Cl_2 (2 mL) was added. The solution was allowed to warm up to −10° C. over 2 h. Sodium hydrogensulfite (10% aqueous solution, 2 mL) was added to reduce the excess of 3-chloroperbenzoic acid. The organic phase separated, diluted with CH_2Cl_2 (30 mL), washed with saturated aqueous Na_2CO_3 (20 mL), water (20 mL), dried over Na_2SO_4 and evaporated to dryness. The residue was dissolved in THF (2.1 mL) and triethylamine trihydrofluoride (0.17 g, 1.1 mmol). The reaction mixture was stirred at room temperature for 18 h. The solution was evaporated to give an oil. The oil was dissolved in ethyl acetate (30 mL) and washed with water (20 mL), 5% aqueous $NaHCO_3$ and brine (20 mL). The organic phase was dried over anhydrous Na_2SO_4 and evaporated. The residue was dissolved in 20% MeOH in water and purified by HPLC on C-18 column (Luna C18, 250×2.12 mm, A=water, B=acetonitrile 20 to 10% B in 65 mL, flow 10 mL min^{-1}, λ260 nm) to give the title compound (0.054 g).

^1H NMR (DMSO-d_6): δ 1.17 (s, 18H), 3.11 (t, 4H, J=6.2 Hz), 3.39 (s, 3H), 3.75 (t, 1H, J=4.8 Hz), 3.934.3 (m, 8H), 5.23 (d, 1H, J=6.4 Hz), 5.8 (d, 1H, J=3.8 Hz), 7.07 (bs, 1H), 7.89 (s, 1H) 7.94 (bs, 1H); ^{13}P NMR (CD_3CN): δ −0.34; MS (AP-ES) m/z 702.00 and 704.00 $[M-H]^-$; HRMS (MALDI) Calcd for $C_{24}H_{39}BrN_3O_{10}PS_2Na$ 726.0890 and 728.0890 found 726.0893 and 728.086.

EXAMPLE 82

3'-Deoxycytidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate]



Step A: N^4-(4,4'-dimethoxytrityl)-3'-deoxycytidine

3'-Deoxycytidine (0.8 g, 3.54 mmol) was mixed with imidazole (2.41 g, 35.4 mmol) and dried over P_2O_5 in vacuum overnight at 40° C. The mixture was dissolved in anhydrous DMF and t-butyldimethylsilyl chloride (2.68 g, 17.78 mmol) was added and the reaction mixture was stirred under an argon atmosphere for 18 h at room temperature. The reaction mixture was diluted with water (100 mL) and extracted with ethyl acetate (2×75 mL). The organic phase was separated, dried over anhydrous Na_2SO_4 and evaporated. The residue was purified by silica gel column chromatography and eluted with ethyl acetate/hexane (6:4) to yield 2',5'-bis(t-butyldimethylsilyl)-3'-deoxycytidine (1.27 g). This was then mixed with DMAP (0.34 g, 2.79 mmol) and dried in vacuum. This mixture was dissolved in anhydrous pyridine (8 mL) and 4,4'-dimethoxytrityl chloride (1.89 g, 5.58 mmol) was added. The reaction mixture was stirred at room temperature under an argon atmosphere for 18 h. Solvent was removed in vacuo. The residue obtained was taken in ethyl acetate (100 mL) and washed with 5% $NaHCO_3$ in water (75 mL) and brine (75 ml). The organic phase was dried over anhydrous Na_2SO_4 and evaporated. The residue obtained was dissolved in THF (28 mL). To this triethylamine trihydrofluoride (2.26 mL, 13.74 mmol) and triethylamine (0.95 mL, 6.87 mmol) were added and stirred at room temperature for 18 h. Solvent was removed and the residue dissolved in ethyl acetate (50 mL), washed with water (50 mL) and 5% $NaHCO_3$ in water (50 mL). The organic phase was dried over anhydrous Na_2SO_4 and evaporated. The residue obtained was purified by silica gel column chromatography and eluted with 5% MeOH in CH_2Cl_2 to yield the title compound (0.66 g).

^{1}H NMR (DMSO-d_6): δ 1.66 (m, 1H), 1.85 (m, 1H1), 3.47 (m, 1H), 3.63 (m, 1H), 3.71 (s, 6H), 4.00 (bs, 1H), 4.19 (m, 1H), 4.96 (t, 1H, J 5.2 Hz), 5.39 (bs, 1H), 5.53 (s, 1H), 6.17 (bs, 1H), 6.83 (d, 4H, J=8.8 Hz), 7.04–7.22 (m, 9H), 7.77 (d, 1H, J=7.6 Hz), 8.27 (bs, 1H); MS (AP-ES) m/z 528.1 $[M-H]^{31}$.

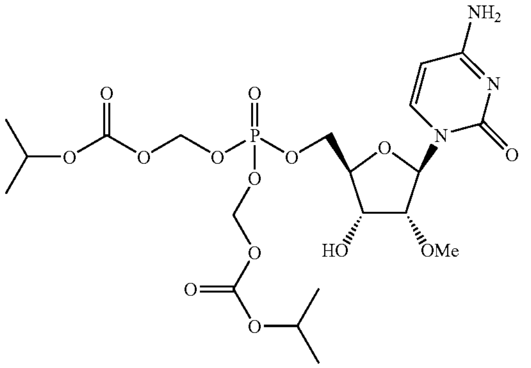
Step B: 3'-Deoxycytidine-5'-[bis-(S-pivaloyl-2-thioethyl) phosphate]

This compound was synthesized following the similar synthetic procedure used for the synthesis of Example 80 starting with N^4-(4,4'-dimethoxytrityl)-3'-deoxycytidine (0.3 g, 0.57 mmol). Other reagents used for the synthesis were 1H-tetrazole (0.04 g, 0.57 mmol), acetonitrile (4 mL), bis-(S-pivaloyl-2-thioethyl)-N,N-diisopropylphosphoramidite (0.52 g, 1.14 mmol) and 3-chloroperbenzoic acid (0.2g, 1.14 mmol, 57–80%) in CH_2Cl_2 (3.6 mL). The product was isolated in 22% yield (0.073 g) after HPLC purification.

^{1}H NMR (200 MHz, DMSO-d_6): δ 1.17 (s, 18H), 1.84 (m, 2H), 3.11 (t, 4H, J=6.4 Hz), 3.93–4.31 (m, 8H), 4.39 (m, 1H), 5.55 (d, 1H, J=4.2 Hz), 5.67(dd, 2H, J=7.4 and 1.8 Hz), 7.1 (bs, 2H), 7.56 (d, 1H, J=7.4 Hz); ^{13}P NMR (CD_3CN): δ −0.71; MS (AP-ES) m/z 596.1 $[M+H]^+$; HRMS (MALDI) Calcd for $C_{23}H_{38}N_3O_9PS_2Na$ 618.1679 found 618.1600.

EXAMPLE 83

2'-O-Methylcytidine-5'-[bis-(isoprovyloxycarbonyloxymethyl)]phosphate



Phosphonomethoxy nucleoside analogs are discussed in C. R. Wagner, V. V. Iyer, and E. J. McIntee, "Pronucleotides: Toward the In Vivo Delivery of Antiviral and Anticancer Nucleotides," Med. Res. Rev., 20: 1–35 (2000), which is incorporated by reference herein in its entirety. They are also disclosed U.S. Pat. Nos. 5,922,695; 5,977,089; 6,043,230; and 6,069,249, the contents of each of which are incorporated by reference herein in their entirety.

Step A: iso-Propyl chloromethyl carbonate

This was prepared according to *Antiviral Chemistry & Chemotherapy* 8: 557 (1997).

Step B: 2'-O-Methylcytidine-5'-phosphate

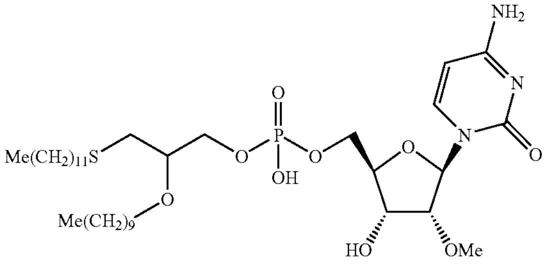
This intermediate was prepared as described in *Tetrahedron Lett.* 50: 5065 (1967).

Step C: 2'-O-Methylcytidine-5'-[bis(isopropyloxycarbonyloxymethyl)]phosphate

2'-O-Methylcytidine-5'-phosphate (0.4 g, 1.19 mmol) was dried over P_2O_5 in vacuum overnight at 40° C. It was then suspended in anhydrous DMF (4 mL). To this mixture was added diisopropylethylamine (0.86 mL, 4.92 mmol) and iso-propyl chloromethyl carbonate (1.56 g, 7.34 mmol). The mixture was heated at 50° C. for 1 h. The reaction mixture was then allowed to come to room temperature. The reaction mixture was stirred at room temperature for 48 h and then filtered. The filtrate was diluted with water (100 mL) and extracted with CH_2Cl_2 (3×50 mL). The organic phase was dried over anhydrous Na_2SO_4 and evaporated. The residue was dissolved in 20% MeOH in water and purified by HPLC on C-18 column (Luna C18,250×2.12 mm, A=water, B=acetonitrile 20 to 10% B in 65 ML, flow 10 mL min^{-1}, λ 260 nm) to give the title compound (2.5 mg). ^{13}P NMR (CD_3CN): δ −3.09; MS (AP-ES) m/z 570.1 $[M+H]^+$.

EXAMPLE 84

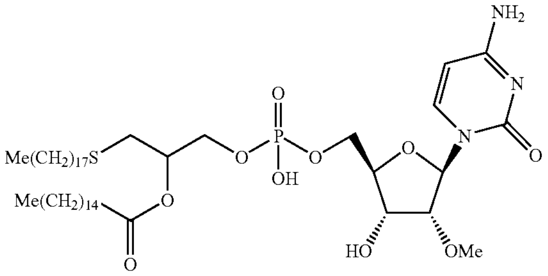
2'-O-Methylcytidine-5'-[(2-decyloxy-3-dodecylthio-1-propyl)phosphate]



The procedure is described for similar nucleoside analogs in German Patent 408366 (1992) and *J. Acquired Immune Defic. Syndr.*2000, 23, 227. The reaction of the appropriately protected 2'-O-methylcytidine with (2-decyloxy-3-dodecylthio-1-propyl)phosphate [prepared by the reaction of 2-decyloxy-3-dodecylthio-1-propanol with $POCl_3$ in ether in presence of triethylamine] under refluxing conditions in a toluene-ether mixture furnishes the desired compound.

EXAMPLE 85

2'-O-Methylcytidine-5'-[rac-(3-octadecylthio-2-palmitoyloxy-1-propyl)phosphate]



This compound is synthesized by the reaction of 2'-O-methylcytidine-5'-monophosphoromorpholidate with rac-1-S-octadecyl-2-O-palmitoyl-1-thioglycerol in pyridine following the similar procedure described for AZT and ddC in *J. Med. Chem.* 39: 1771 (1996).

EXAMPLE 86

Nucleoside 5'-Triphosphates

The nucleoside 5'-triphosphates of the present invention were prepared according to the general procedures described in *Chem. Rev.*100: 2047 (2000).

EXAMPLE 87

Purification and Purity Analysis of Nucleoside 5'-Triphosphates

Triphosphates were purified by anion exchange (AX) chromatography using a 30×100 mm Mono Q column (Pharmacia) with a buffer system of 50 mM Tris, pH 8. Elution gradients were typically from 40 mM NaCl to 0.8 M NaCl in two column volumes at 6.5 mL/min. Appropriate fractions from anion exchange chromatography were collected and desalted by reverse-phase (RP) chromatography using a Luna C18 250×21 mm column (Phenomenex) with a flow rate of 10 ml/min. Elution gradients were generally from 1% to 95% methanol in 14 min at a constant concentration of 5 mM triethylammonium acetate (TEAA).

Mass spectra of the purified triphosphates were determined using on-line HPLC mass spectrometry on a Hewlett-Packard (Palo Alto, Calif.) MSD 1100. A Phenomenex Luna (C18(2)), 150×2 mm, plus 30×2 mm guard column, 3-μm particle size was used for RP HPLC. A 0 to 50% linear gradient (15 min) of acetonitrile in 20 mM TEAA (triethylammonium acetate) pH 7 was performed in series with mass spectral detection in the negative ionization mode. Nitrogen gas and a pneumatic nebulizer were used to generate the electrospray. The mass range of 150–900 was sampled. Molecular masses were determined using the HP Chemstation analysis package.

The purity of the purified triphosphates was determined by analytical RP and AX HPLC. RP HPLC with a Phenomenex Luna or Jupiter column (250×4.6 mm), 5-μm particle size was typically run with a 2–70% acetonitrile gradient in 15 min in 100 mM TEAA, pH 7. AX HPLC was performed on a 1.6×5 mm Mono Q column (Pharmacia). Triphosphates were eluted with a gradient of 0 to 0.4 M NaCl at constant concentration of 50 mM Tris, pH 8. Purity of the triphosphates was generally >80%.

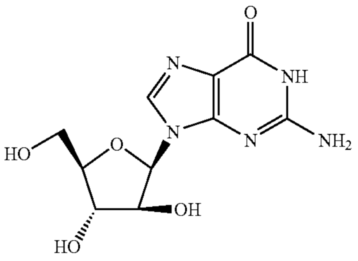
EXAMPLE 88

Nucleoside 5'-Monophosphates

The nucleoside 5'-monophosphates of the present invention were prepared according to the general procedure described in *Tetrahedron Lett.* 50: 5065 (1967).

EXAMPLE 89

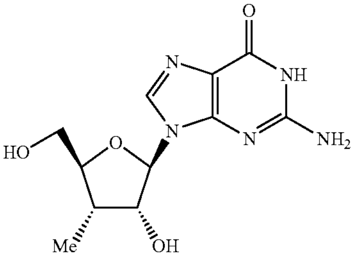
2-Amino-9-(β-D-arabinofuranosyl)-9H-purin-6(1H)-one



This compound was obtained from commercial sources.

EXAMPLE 90

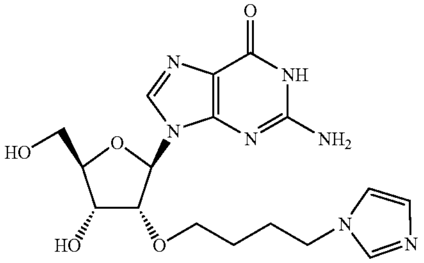
3'-Deoxy-3'-methylguanosine



This compound was prepared following procedures described in U.S. Pat. No. 3,654,262 (1972).

EXAMPLE 91

2'-O-[4-(Imidazolyl-1)butyl]guanosine



Step A: 2'-O-[4-(Imidazolyl-1)butyl]-2-aminoadenosine

A solution 2-aminoadenosine (7.36 g, 26 mmol) in dry DMF (260 mL) was treated portionwise with 60% NaH (3.92 g, 1000 mmol). After 1 hr., a solution of bromobutylimidazole (9.4 g, 286 mmol) in DMF (20ml) was added. After 16 hrs., the solution was conc. in vacuo, partitioned between H_2O/EtOAc and separated. The aqueous layer was evaporated, and the residue was chromatographed on silica gel ($CHCl_3$/MeOH) to afford the title nucleoside as a white solid; yield 4.2 g.

^{1}H NMR (DMSO-d_6): δ 1.39 (t, 2H), 1.67 (t, 2H), 3.3–3.7 (m, 4H), 3.93 (m, 3H), 4.29 (m, 2H), 4.40 (d, 1H), 5.50 (5, 1H), 5.72 (d, 1H), 5.82 (bs, 2H), 6.72 (bs, 2H), 6.86 (s, 1H), 7.08 (s, 1H), 7.57 (s, 1H). 7.91 (s, 1H).

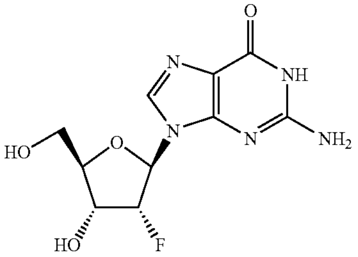
Step B: 2'-O-[4-(Imidazolyl-1)butyl]guanosine

A mixture of the intermediate from Step A (3.2 g, 8 mmol) in H_2O (200 mL), DMSO (10 mL), trisodium phosphate (10 g), and adenosine deaminase (0.3 g) was stirred at room temperature and pH 7. The solution was filtered and and then evaporated. The resulting solid was crystallized from EtOAc/MeOH to afford the title compound as a white solid; yield 2.6 g.

^{1}H NMR (DMSO-d_6): δ 1.39 (t, 2H), 1.67 (t, 2H), 3.3–3.7 (m, 4H), 3.93 (m, 3H), 4.29 (m, 2H), 5.10 (t, 1H), 5.20 (d, 1H), 5.79 (d, 1H), 6.50 (bs, 2H), 6.86 (s, 1H), 7.08 (s, 1H), 7.57 (s, 1H) 7.9 (s, 1H).

EXAMPLE 92

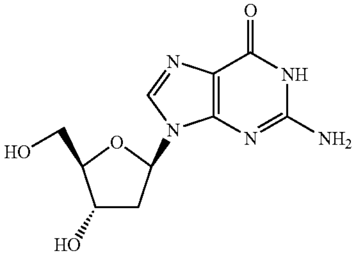
2'-Deoxy-2'-fluoroguanosine



This compound was prepared following the conditions described in *Chem. Pharm. Bull*. 29: 1034 (1981).

EXAMPLE 93

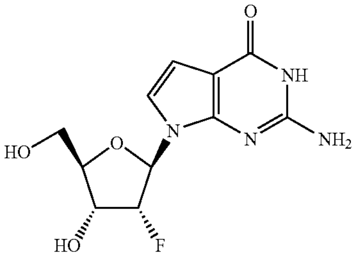
2'-Deoxyguanosine



This compound was obtained from commercial sources.

EXAMPLE 94

2-Amino-7-(2-deoxy-2-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



Step A: 2-Amino-4-chloro-7-(2,3,5-tri-O-benzyl-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]-pyrimidine

To a suspension of 2-amino-4-chloro-1H-pyrrolo[2,3-d] pyrimidine [*Liebigs Ann. Chem*. 1: 137 (1983)] (3.03 g, 18 mmol) in anhydrous MeCN (240 mL), powdered KOH (85%; 4.2 g, 60 mmol) and tris[2-(2-methoxyethoxy)-ethyl] amine (0.66 mL, 2.1 mmol) were added and the mixture was stirred at room temperature for 10 min. Then a solution of 2,3,5-tri-O-benzyl-D-arabinofuranosyl bromide [prepared from corresponding 1-O-p-nitrobenzoate (11.43 g, 20.1 mmol) according to Seela et al., *J. Org. Chem*. (1982), 47, 226] in MeCN (10 mL) was added and stirring continued for another 40 min. Solid was filtered off, washed with MeCN (2×25 mL) and combined filtrate evaporated. The residue was purified on a silica gel column with a solvent system of hexanes/EtOAc: 7/1, 6/1 and 5/1. Two main zones were separated. From the more rapidly migrating zone was isolated the α anomer (0.74 g) and from the slower migrating zone the desired β anomer (4.01 g).

Step B: 2-Amino-7-(β-D-arabinofuranosyl)-4-chloro-7H-pyrrolo[2,3-d]pyrimidine

To a solution of the compound from Step A (4.0 g, 7 mmol) in CH_2Cl_2 (150 ml) at −78° C. was added a solution of 1.0 M BCl_3 in CH_2Cl_2 (70 mL, 70 mmol) during 45 min. The mixture was stirred at −78° C. for 3 h and at −20° C. for 2.5 h. MeOH—CH_2Cl_2 (70 mL, 1:1) was added to the mixture, which was then stirred at −20° C. for 0.5 h and neutralized with conc. aqueous NH_3 at 0° C. The mixture was stirred at room temperature for 10 min. and then filtered. The solid was washed with MeOH—CH_2Cl_2 (70 mL, 1:1) and the combined filtrate evaporated. The residue was purified on a silica gel column with a solvent system of CH_2Cl_2/MeOH: 20/1 to give the desired nucleoside (1.18 g) as a white solid.

Step C: 2-Amino-7-[3,5-O-(1,3,3-tetraisopropyldisiloxane-1,3-diyl)-β-D-arabinofuranosyl]-4-chloro-7H-pyrrolo[2,3-d]pyrimidine

The compound from Step B (0.87 g, 2.9 mmol) and imidazole (0.43 g, 5.8 mmol were dissolved in DMF (3.5 mL). 1,3-Dichloro-1,1,3,3-tetraisopropyldisiloxane (1.0 mL) was added to the solution. The reaction mixture was stirred at room temperature for 1 h and then evaporated. The residue was partitioned between CH_2Cl_2 (150 mL) and water (30 mL). The layers were separated. The organic layer was dried (Na_2SO_4) and evaporated. The residue was purified on a silica gel column with a solvent system of hexanes/EtOAc: 7/1 and 5/1 to give the title compound (1.04 g).

Step D: 2-Amino-7-[2-O-acetyl-3,5-0-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)-β-D-arabinofuranosyl]-4-chloro-7H-pyrrolo[2,3-d]pyrimidine

A mixture of the compound from Step C (0.98 g, 1.8 mmol) in MeCN (12 mL), Et_3N (0.31 mL) Ac_2O (0.21 mL) and DMAP (5 mg, 0.25 eq.) was stirred at room temperature for 5 h and then evaporated. The oily-residue was dissolved in EtOAc (200 mL), washed with water (2×20 mL), dried (Na_2SO_4) and evaporated to yield pure title compound (1.12 g).

Step E: 2-Amino-7-[2-O-acetyl-β-D-arabinofuranosyl]-4-chloro-7H-pyrrolo[2,3-d]pyrimidine

To an ice-cold solution of the compound from Step D (0.95 g, 1.63 mmol) in THF (10 mL) and AcOH (0.19 mL) was added dropwise 1.0 M tetrabutylammonium fluoride solution in THF (3.4 mL) and stirred at 0° C. for 15 min. The solution was concentrated and the oily residue applied onto a silica gel column packed in CH_2Cl_2 and eluted with CH_2Cl_2/MeOH: 50/1, 25/1 and 20/1. Appropriate fractions were pooled and evaporated to give the title nucleoside (0.56 g) as a white solid.

Step F: 2-Amino-7-[2-O-acetyl-3,5-di-O-(tetrahydro-2-pyranyl)-β-D-arabinofuranosyl]-4-chloro-7H-pyrrolo[2,3-d]pyrimidine

To a solution of the compound from Step E (0.5 g, 1.46 mmol) in CH_2Cl_2 (10 mL) and 3,4-dihydro-2-H-pyrane (0.67 mL) was added dropwise TMSI (30 μL, 0.2 mmol). The reaction mixture was stirred at room temperature for 1 h and then evaporated. The oily residue was purified on a silica gel column packed in a solvent system of hexanes/EtOAc/Et_3N: 75/25/1 and eluted with a solvent system of hexanes/EtOAc: 3/1. The fractions containing the product were collected and evaporated to give the desired compound (0.60 g).

Step G: 2-Amino-7-[3,5-di-O-(tetrahydro-2-pyranyl)-β-D-arabinofuranosyl]-4-chloro-7H-pyrrolo[2,3-d]pyrimidine

A mixture of the compound from Step F (0.27 g, 0.53 mmol) and methanolic ammonia (saturated at 0° C.; 10 mL) was kept overnight at 0° C. Evaporation of the solvent yielded the desired compound (0.25 g).

Step H: 2-Amino-7-[2-deoxy-2-fluoro-3,5-di-O-(tetrahydro-2-pyranyl)-β-D-ribofuranosyl]-4-chloro-7H-pyrrolo[2,3-d]pyrimidine

To a solution of the compound from Step G (0.24 g, 0.51 mmol) in CH_2Cl_2 (5 mL) and pyridine (0.8 mL) at −60° C. was added diethylaminosulfur trifluoride (DAST; 0.27 mL) dropwise under Ar. The solution was stirred at −60° C. for 0.5 h, at 0° C. overnight and at room temperature for 3 h. The mixture was diluted with CH_2Cl_2 (25 mL) and poured into saturated aqueous $NaHCO_3$ (15 mL). The organic layer was washed with water (10 mL), dried (Na_2SO_4) and evaporated. The residue was purified on a silica gel column with a solvent system of hexanes/EtOAc: 5/1 to give the title compound (45 mg) as a pale yellow foam.

Step I: 2-Amino-7-(2-deoxy-2-fluoro-β-D-ribofuranosyl)-4-chloro-7H-pyrrolo[2,3-d]-primidine

A solution of the compound from Step H (40 mg. 0.08 mmol) in EtOH (2 mL) was stirred with pyridinium p-toluenesulfonate (40 mg, 0.16 mmol) at 60° C. for 3 h. The mixture was then evaporated and the residue purified on a silica gel column with a solvent system of hexanes/EtOAc: 1/1 and 1/2 to give the desired compound (24 mg).

Step J: 2-Amino-7-(2-deoxy-2-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one

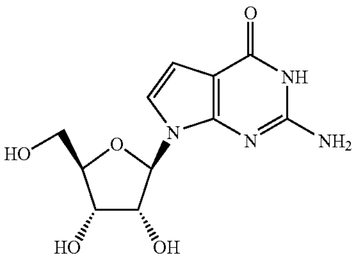
A mixture of the compound from Step I (4 mg, 0.08 mmol) in 2N aqueous NaOH (1.2 mL) was stirred at reflux temperature for 1.5 h. The solution was cooled in an ice-bath, neutralized with 2 N aqueous HCl and evaporated to dryness. The residue was suspended in MeOH, mixed with silica gel and evaporated. The solid residue was placed onto a silica gel column (packed in a solvent system of CH_2Cl_2/MeOH: 10/1) which was eluted with a solvent system of CH_2Cl_2/MeOH: 10/1. The fractions containing the product were collected and evaporated to dryness to yield the title compound (20 mg) as a white solid.

^1H NMR (CD_3OD): δ 3.73, 3.88 (2dd, 2H, J=12.4, 3.8, 2.6 Hz), 4.01 (m, 1H), 4.47 (ddd, 1H J=16.5, 6.6 Hz), 5.14 (ddd, 1H, J=5.3, 4.7 Hz), 6.19 (dd, 1H, J=17.8, 3.0 Hz), 6.39 (d, 1H, J=3.6 Hz), 6.95 (d, 1H).

^{19}F NMR (CD_3OD): δ −206.53 (dt).

EXAMPLE 95

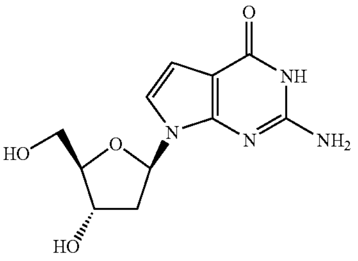
2-Amino-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



This compound was prepared following the procedures described in *J. Chem. Soc. Perkin Trans.* 1, 2375 (1989).

EXAMPLE 96

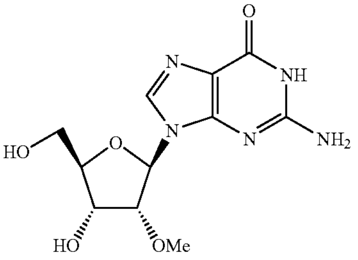
2-Amino-7-(2-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



This compound was prepared following the procedures in *Tetrahedron Lett*. 28: 5107 (1987).

EXAMPLE 97

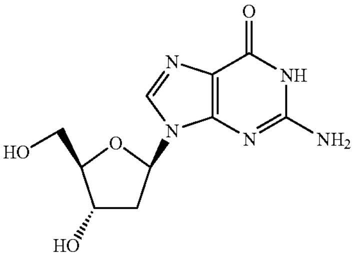
6-Amino-1-(2-O-methyl-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one



This compound was prepared in a manner similar to the preparation of 2-amino-7-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyridin-4(3H)-one (Example 23).

EXAMPLE 98

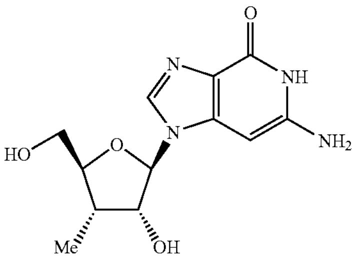
6-Amino-1-(2-deoxy-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one



This compound was prepared following the procedures described in *J. Med. Chem*. 26: 286 (1983).

EXAMPLE 99

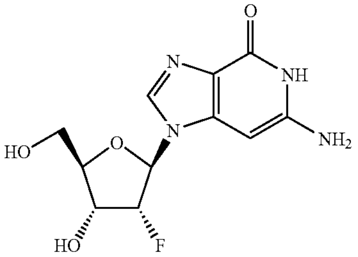
6-Amino-1-(3-deoxy-3-methyl-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one



This compound was prepared in a manner similar to the preparation of 2-amino-7-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one (Example 23).

EXAMPLE 100

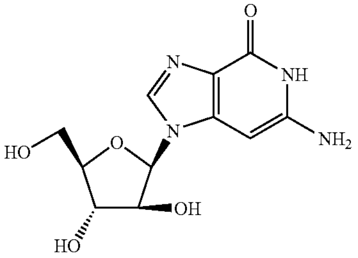
6-Amino-1-(2-deoxy-2-fluoro-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one



This compound was prepared in a manner similar to the preparation of 2-amino-7-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one (Example 23).

EXAMPLE 101

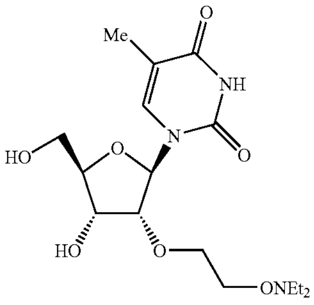
6-Amino-1-(β-D-arabinofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one



A preparation of this compound is given in Eur. Pat. Appln. 43722 A1 (1982).

EXAMPLE 102

2'-O-[2-(N,N-diethylaminooxy)ethyl]-5-methyluridine



Step A: 5'-O-tert-Butyldiphenylsilyl-2'-O-(2-hydroxyethyl)-5-methyluridine

In a 2 L stainless steel, unstirred pressure reactor was added borane in tetrahydrofuran (1.0 M, 2.0 eq, 622 mL). In the fume hood and with manual stirring, ethylene glycol (350 mL, excess) was added cautiously at first until the evolution of hydrogen gas subsided. 5'-O-tert-Butyldiphenylsilyl-O^2-2'-anhydro-5-methyluridine (149 g, 0.311 mol) and sodium bicarbonate (0.074 g) were added with manual stirring. The reactor was sealed and heated in an oil bath until an internal temperature of 160° C. was reached and then maintained for 16 h(pressure <100 psig). The reaction vessel was cooled to ambient and opened. The reaction mixture was concentrated under reduced pressure (10 to 1 mm Hg) in a warm water bath (40–100° C.) with the more extreme conditions used to remove the ethylene glycol. The residue was purified by column chromatography (2 kg silica gel, ethyl acetate:hexanes gradient 1:1 to 4:1). The appropriate fractions were combined, stripped and dried to product as white crisp foam (84 g), contaminated starting material (17.4 g) and pure reusable starting material (20 g). TLC and NMR were consistent with 99% pure product.

^{1}H NMR (DMSO-d_6): δ 1.05 (s, 9H), 1.45 (s, 3H), 3.5–4.1 (m, 8H), 4.25 (m, 1H), 4.80 (t, 1H), 5.18 (d, 2H), 5.95 (d, 1H), 7.35–7.75 (m, 11H), 11.42 (s, 1H).

Step B: 2'-O-[2-(2-phthalimidoxy)ethyl]-5'-t-butyldiphenylsilyl-5-methyluridine

5'-O-tert-Butyldiphenylsilyl-2'-O-(2-hydroxyethyl)-5-methyluridine (20 g, 36.98 mmol) was mixed with triphenylphosphine (11.63 g, 44.36 mmol) and N-hydroxyphthalimide (7.24 g, 44.36 mmol). It was then dried over P_2O_5 under high vacuum for two days at 40° C. The reaction mixture was flushed with argon and dry THF (369.8 mL) was added to get a clear solution. Diethyl azodicarboxylate (6.98 mL, 44.36 mmol) was added dropwise to the reaction mixture. The rate of addition was maintained such that resulting deep red coloration is just discharged before adding the next drop. After the addition was complete, the reaction was stirred for 4 h. By that time TLC showed the completion of the reaction (ethyl acetate/hexane, 60:40). The solvent was evaporated under vacuum. Residue obtained was placed on a flash silica gel column and eluted with ethyl acetate-hexane (60:40) to give the title compound as a white foam (21.8.g).

^{1}H NMR (DMSO-d_6): δ 11.32 (s, 1H), 7.82 (m, 4H), 7.6–7.65 (m, 5H), 7.34–7.46 (m, 6H), 5.90 (d, 1H, J=6Hz), 5.18 (d, J=5.6 Hz), 4.31 (bs, 2H), 4.25 (m, 1H), 4.09 (t, 1H, J=5.6 Hz), 3.81–3.94 (m, 5H), 1.44 (s, 3H), 1.1 (s, 9H); ^{13}C NMR ($CDCl_3$): δ 11.8, 19.40, 26.99, 62.62, 68.36, 68.56, 77.64, 83.04, 84.14, 87.50, 110.93, 123.59, 127.86, 129.89, 132.45, 134.59, 134.89, 135.17, 150.50, 163.63, 163.97; MS [FAB] m/z 684 $[M-H]^-$.

Step C: 5'-O-tert-Butyldiphenylsilyl-2'-O-[2-(acetaldoximinooxy)ethyl]-5-methyluridine

2'-O-[2-(2-Phthalimidoxy)ethyl]-5'-t-butyldiphenylsilyl-5-methyluridine (10 g, 14.6 mmol) was dissolved in CH_2Cl_2 (146 mL) and cooled to −10° C. in an isopropanol-dry ice bath. To this methylhydrazine (1.03 mL, 14.6 mmol) was added dropwise. Reaction mixture was stirred at −10° C. to 0° C. for 1 h. A white precipitate formed and was filtered and washed thoroughly with CH_2Cl_2 (ice cold). The filtrate was evaporated to dryness. Residue was dissolved in methanol (210 mL) and acetaldehyde (0.89 mL, 16 mmol) was added and stirred at room temperature for 12 h. Solvent was removed in vacuo and residue was purified by silica gel column chromatography using and ethyl acetate/hexane (6:4) as solvent system to yield the title compound (4.64 g).

^1H NMR (DMSO-d_6): δ 1.02 (s, 9H), 1.44 (s, 3H), 1.69 (dd, 3H, J=5.6 Hz), 3.66 (m, 1H), 3.76 (m, 2H), 3.94 (m, 2H), 4.05 (s, 2H), 4.15 (m, 1H), 4.22 (m, 1H), 5.18 (d, 1H, J=6.0 Hz), 5.9 (dd, 1H, J=4.4 Hz), 7.36 (m, 1H), 7.40 (m, 7H), 7.63 (m, 5H), 11.38 (s, 1H), ^{13}C NMR ($CDCl_3$): δ 11.84, 15.05, 19.38, 26.97, 63.02, 68.62, 70.26, 71.98, 72.14, 82.72, 84.34, 87.02, 111.07, 127.89, 130.02, 134.98, 135.13, 135.42, 147.85, 150.51, 164.12; HRMS (FAB) Calcd for $C_{30}H_{39}N_3O_7$ $SiNa^{\oplus}$ 604.2455, found 604.2471.

Step D: 5'-O-tert-Butyldiphenylsilyl-2'-O-[2-(N N-diethylaminooxy)ethyl]-5-methyluridine

5'-O-tert-Butyldiphenylsilyl-2'-O-[2-(acetaldoximinooxy)ethyl]-5-methyluridine (4.5 g, 7.74 mmol) was dissolved in 1M pyridinium p-toluenesulfonate (PPTS) in MeOH (77.4 mL). It was then cooled to 10° C. in an ice bath. To this mixture $NaBH_3CN$ (0.97 g, 15.5 mmol) was added and the mixture was stirred at 10° C. for 10 minutes. Reaction mixture was allowed to come to room temperature and stirred for 4 h. Solvent was removed in vacuo to give an oil. Diluted the oil with ethyl acetate (100 mL), washed with water (75 mL), 5% $NaHCO_3$ (75 mL) and brine (75 mL). The organic phase was dried over anhydrous Na_2SO_4 and evaporated. Residue obtained was dissolved in 1M PPTS in MeOH (77.4 mL), acetaldehyde (0.48 mL, 8.52 mmol) was added and stirred at ambient temperature for 10 minutes. Then reaction mixture was cooled to 10° C. in an ice bath and $NaBH_3CN$ (0.97 g, 15.50 mmol) was added and stirred at 10° C. for 10 minutes. Reaction mixture was allowed to come to room temperature and stirred for 4 h. Solvent was removed in vacuo to get an oil. The oil was dissolved in ethyl acetate (100 mL), washed with water (75 mL), 5% $NaHCO_3$ (75 mL) and brine (75 mL). The organic phase was dried over anhydrous Na_2SO_4 and evaporated to dryness. The residue obtained was purified by silica gel column chromatography and eluted with CH_2Cl_2/MeOH /NEt_3, 94:5:1 to give title compound (3.55 g) as a white foam.

^1H NMR (DMSO-d_6): δ 0.95 (t, 6H, J=7.2 Hz), 1.03 (s, 9H), 1.43 (s, 3H), 2.58 (q, 4H, J=7.2 Hz), 3.59 (m, 1H), 3.73 (m, 3H), 3.81 (m, 1H), 3.88 (m, 1H), 3.96 (m, 2H), 4.23 (m, 1H), 5.21 (d, 1H, J=5.6 Hz), 5.95 (d, 1H, J=6.4 Hz), 7.43 (m, 7H), 7.76 (m, 4H), 11.39 (s, 1H); ^{13}C NMR ($CDCl_3$): δ 11.84, 19.35, 26.97, 52.27, 63.27, 68.81, 70.27, 72.27, 82.64, 84.47, 86.77, 111.04, 127.87, 130.01, 135.11, 135.41, 141.32, 150.48, 164.04; HRMS (FAB), Calcd for $C_{32}H_{45}N_3O_7SiCs^{\oplus}$, 744.2081, found 744.2067.

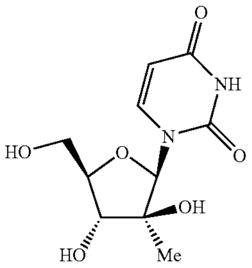
Step E: 2'-O-[2-(N,N-diethylaminooxy)ethyl]-5-methyluridine

A mixture of triethlyamine trihydrogenfluoride (4.39 mL, 26.81 mmol) and triethylamine (1.87 mL, 13.41 mmol) in THF (53.6 mL) was added to 5'-O-tert-butyldiphenylsilyl-2'-O-[2-(N,N-diethylaminooxy)ethyl]-5-methyluridine (3.28 g, 5.36 mmol). The reaction mixture was stirred at room temperature for 18 h. Solvent was removed in vacuo. The residue was placed on a silica gel column and eluted with CH_2Cl_2/MeOH/NEt_3, 89: 10: 1, to yield the title compound (1.49 g).

^1H NMR (DMSO-d_6): δ 0.97 (t, 6H, J=7.2 Hz), 1.75 (s, 3H), 2.58 (q, 4H, J=7.2 Hz), 3.55 (m, 4H), 3.66 (m, 2H), 3.83 (bs, 1H), 3.95 (t, 1H, J=5.6 Hz), 4.11 (q, 1H, J=4.8 Hz and 5.6 Hz), 5.05 (d, 1H, J=5.6 Hz), 5.87 (d, 1H, J=6.0 Hz), 7.75 (s, 1H), 11.31 (s, 1H); ^{13}C NMR ($CDCl_3$): 5 11.75, 12.27, 52.24, 61.31, 68.86, 70.19, 72.25, 81.49, 85.10, 90.29, 110.60, 137.79, 150.57, 164.37; HRMS (FAB) Calcd for $C_{16}H_{28}N_3O_7^{\oplus}$ 374.1927, found 374.1919.

EXAMPLE 103

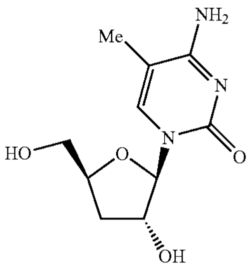
1-(2-C-Methyl-β-D-arabinofuranosyl)uracil



This compound was prepared following the procedures described in *Chem. Pharm. Bull.* 35: 2605 (1987).

EXAMPLE 104

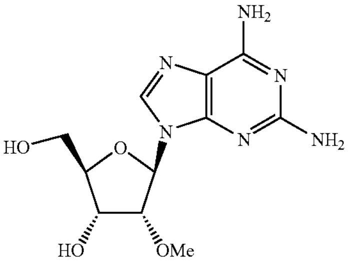
5-Methyl-3'-deoxycytidine



This compound was prepared following the procedures described in *Chem. Pharm. Bull.* 30: 2223 (1982).

EXAMPLE 105

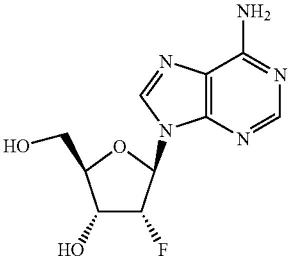
2-Amino-2'-O-methyladenosine



This compound was obtained from commercial sources.

EXAMPLE 106

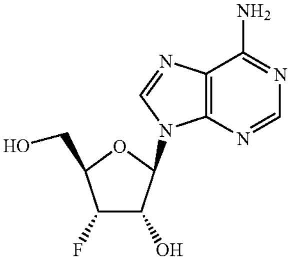
2'-Deoxy-2'-fluoroadenosine



This compound was obtained from commercial sources.

EXAMPLE 107

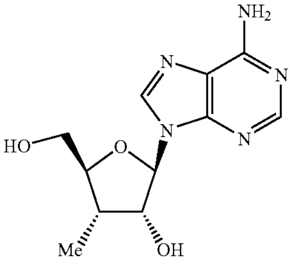
3'-Deoxy-3'-fluoroadenosine



This compound was prepared following the procedures described in *Nucleosides Nucleotides* 10: 719 (1991).

EXAMPLE 108

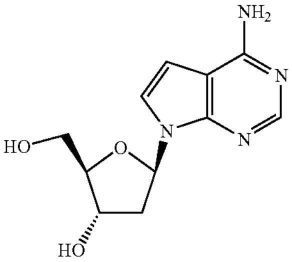
3'-Deoxy-3'-methyladenosine



This compound was prepared following the procedures described in *J. Med. Chem.* 19: 1265 (1976).

EXAMPLE 109

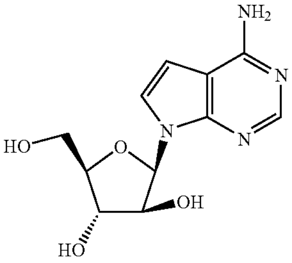
2-Amino-7-(2-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2.3-d]pyrimidine



This compound was prepared following the procedures described in *J. Am. Chem. Soc.* 106: 6379 (1984).

EXAMPLE 110

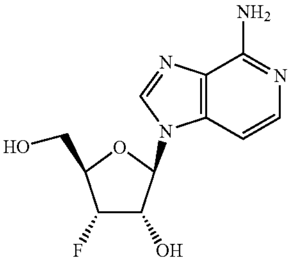
4-Amino-7-(β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



This compound is described in U.S. Pat. No. 4,439,604, which is incorporated by reference herein in its entirety.

EXAMPLE 111

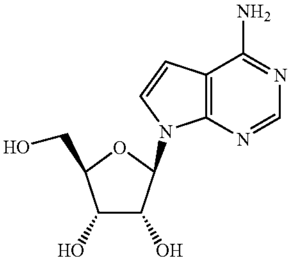
4-Amino-1-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridine



This compound can be prepared readily by the similar method described for the preparation of Example 24 except the nucleobase is 3-deazaadenine.

EXAMPLE 112

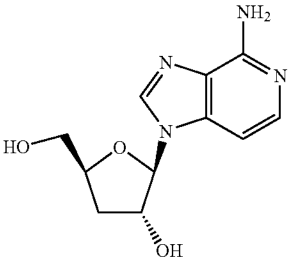
4-Amino-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine (tubercidin)



This compound was obtained from commercial sources.

EXAMPLE 113

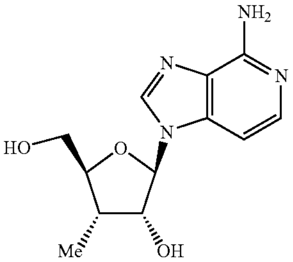
4-Amino-1-(3-deoxy-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridine



This compound is described in *Acta Crystallogr., Sect. C: Cryst. Struct. Commun.* C43: 1790 (1987).

EXAMPLE 114

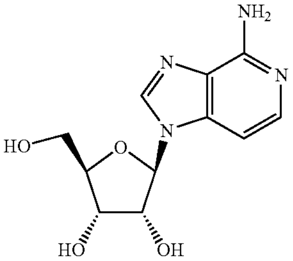
4-Amino-1-(3-deoxy-3-methyl-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridine



The procedure described earlier for Example 23 is used to synthesize this example by reacting the appropriately substituted 3-C-methyl-sugar intermediate with a protected 3-deazaadenine derivative.

EXAMPLE 115

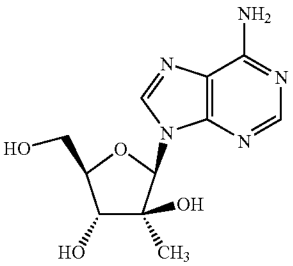
4-Amino-1-β-D-ribofuranosyl-1H-imidazo[4,5-c]pyridine



This compound was obtained from commercial sources.

EXAMPLE 116

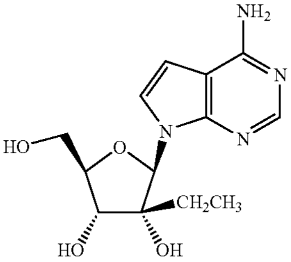
9-(2-C-Methyl-β-D-arabinofuranosyl)adenine



This compound is prepared from 4-amino-9-(3,5-bis-O-tert-butyldimethylsilyl-β-D-erythro-pentofuran-2-ulosyl)purine (*J. Med. Chem.* 1992, 35, 2283) by reaction with MeMgBr and deprotection as described in Example 61.

EXAMPLE 117

4-Amino-7-(2-C-ethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



Step A: 3,5-Bis-O-(2,4-dichlorophenylmethyl)-2-C-ethyl-1-O-methyl-α-D-ribofuranose

To diethyl ether (300 mL) at −78° C. was slowly added EtMgBr (3.0 M, 16.6 mL) and then dropwise the compound from Step B of Example 62 (4.80 g, 10.0 mmol) in anhydrous Et_2O (100 mL). The reaction mixture was stirred at −78° C. for 15 min, allowed to warm to −15° C. and stirred for another 2 h, and then poured into a stirred mixture of water (300 mL) and Et_2O (600 mL). The organic phase was separated, dried (MgSO4), and evaporated in vacuo. The crude product was purified on silica gel using ethyl acetate/hexane (1:2) as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (3.87 g) as a colorless oil.

Step B: 4-Chloro-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C-ethyl-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine

To a solution of the compound from Step A (1.02 mg, 2.0 mmol) in dichloromethane (40 mL) was added HBr (5.7 M in acetic acid) (1.75 mL, 10.0 mmol) dropwise at 0° C. The resulting solution was stirred at rt for 2 h, evaporated in vacuo and co-evaporated twice from toluene (10 mL). The oily residue was dissolved in acetonitrile (10 mL) and added to a vigorously stirred mixture of 4-chloro-1H-pyrrolo[2,3-d]pyrimidine (307 mg, 2.0 mmol), potassium hydroxide (337 mg, 6.0 mmol) and tris[2-(2-methoxyethoxy)ethyl]amine (130 mg, 0.4 mmol) in acetonitrile (10 mL). The resulting mixture was stirred at room temperature overnight, and then poured into a stirred mixture of saturated ammonium chloride (100 mL) and ethyl acetate (100 mL). The organic layer was separated, washed with brine (100 mL), dried over $MgSO_4$, filtered and evaporated in vacuo. The crude product was purified on silica gel using ethyl acetate/hexane (1:2) as eluent to give the desired product (307 mg) as a colorless foam.

Step C: 4-Chloro-7-(2-C-ethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

To a solution of the compound from Step B (307 mg, 0.45 mmol) in dichloromethane (8 mL) was added boron trichloride (1M in dichloromethane) (4.50 mL, 4.50 mmol) at −78° C. The mixture was stirred at −78° C. for 1 h, then at −10° C. for 3 h. The reaction was quenched by addition of methanol/dichloromethane (1:1) (10 mL), stirred at −15° C. for 30 min, and neutralized by addition of aqueous ammonium hydroxide. The mixture was evaporated in vacuo and the resulting oil purified on silica gel using methanol/dichloromethane (1:9) as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (112 mg) as a colorless foam.

Step D: 4-Amino-7-(2-C-ethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

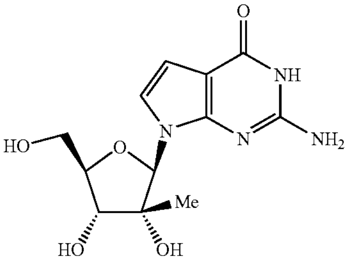
To the compound from Step C (50 mg, 0.16 mmol) was added saturated ammonia in methanol (4 mL). The mixture was stirred at 75° C. for 72 h in a closed container, cooled and evaporated in vacuo. The crude mixture was purified on silica gel using methanol/dichloromethane (1:9) as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (29 mg) as a colorless powder.

^1H NMR (200 MHz, DMSO-d_6): δ 0.52 (t, 3H), 1.02 (m, 2H), 4.01–3.24 (m, 6H), 5.06 (m, 1H), 6.01 (s, 1H), 6.51 (d, 1H), 6.95 (s br, 2H), 6.70 (d, 1H), 7.99 (s, 1H).

LC-MS: Found: 295.2 (M+H^+); calc. for $C_{13}H_{18}N_4O_4$+H^+: 295.14.

EXAMPLE 118

2-Amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



Step A: 2-Amino-4-chloro-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

To an ice-cold solution of product from Step C of Example 62 (1.27 g, 2.57 mmol) in CH_2Cl_2 (30 mL) was added HBr (5.7 M in acetic acid; 3 mL) dropwise. The reaction mixture was stirred at room temperature for 2 h, concentrated in vacuo and co-evaporated with toluene (2×15 mL). The resulting oil was dissolved in MeCN (15 mL) and added dropwise into a well-stirred mixture of 2-amino-4-chloro-7H-pyrrolo[2,3-d]pyrimidine [for preparation see *Heterocycles* 35: 825 (1993)] (433 mg, 2.57 mmol), KOH (85%, powdered) (0.51 g, 7.7 mmol), tris-[2-(2-methoxyethoxy)ethyl]amine (165 μL, 0.51 mmol) in acetonitrile (30 mL). The resulting mixture was stirred at rt for 1 h, filtered and evaporated. The residue was purified on a silica gel column using hexanes/EtOAc, 5/1, 3/1 and 2/1 as eluent to give the title compound as a colorless foam (0.65 g).

Step B: 2-Amino chloro-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

To a solution of the product from Step A (630 mg, 1.0 mmol) in CH_2Cl_2 (20 mL) at −78° C. was added boron trichloride (1M in CH_2Cl_2) (10 mL, 10 mmol). The mixture was stirred at −78° C. for 2 h, then at −20° C. for 2.5 h. The reaction was quenched with CH_2Cl_2/MeOH (1:1) (10 mL), stirred at −20° C. for 0.5 h, and neutralized at 0° C. with aqueous ammonia. The solid was filtered, washed with CH_2Cl_2/MeOH (1:1) and the combined filtrate evaporated in vacuo. The residue was purified on a silica gel column with CH_2Cl_2/MeOH, 50/1 and 20/1 as eluent to give the title compound as a colorless foam (250 mg).

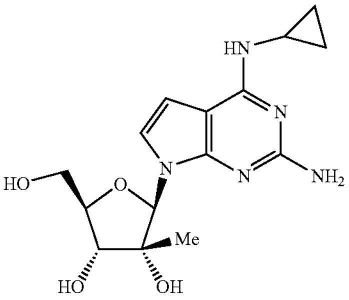
Step C: 2-Amino-7-(2-C-methyl-β-D-ribofiuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one

A mixture of product from Step B (90 mg, 0.3 mmol) in aqueous NaOH (2N, 9 mL) was heated at reflux temperature for 5 h, then neutralized at 0° C. with 2 N aqueous HCl and evaporated to dryness. Purification on a silica gel column with CH_2Cl_2/MeOH, 5/1 as eluent afforded the title compound as a white solid (70 mg).

^{1}H NMR (200 MHz, CD_3OD): δ 0.86 (s, 3H), 3.79 (m 1H), 3.90–4.05 (m, 3H), 6.06 (s, 1H), 6.42 (d, J=3.7 Hz, 1H), 7.05 (d, J=3.7 Hz, 1H).

EXAMPLE 119

2-Amino-4-cyclopropylamino-7-(2-C-methyl-β-D-ribofiuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

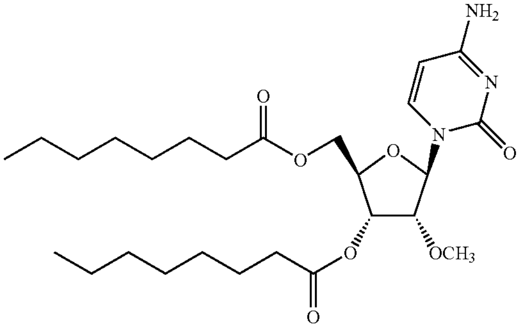


A solution of 2-amino-4-chloro-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine (Example 118, Step B) (21 mg, 0.07 mmol) in cyclopropylamine (0.5 mL) was heated at 70° C. for two days, then evaporated to an oily residue and purified on a silica gel column with CH_2Cl_2/MeOH, 20/1, as eluent to give the title compound as a white solid (17 mg).

^{1}H NMR (200 MHz, CD_3CN): δ 0.61 (m, 2H), 0.81 (m, 2H), 0.85 (s, 3H), 2.83 (m, 1H), 3.74–3.86 (m, 1H), 3.93–4.03 (m, 2H), 4.11 (d, J=8.9 Hz, 1H), 6.02 (s, 1H), 6.49 (d, J=3.7 Hz, 1H), 7.00 (d, J=3.7 Hz, 1H).

EXAMPLE 120

3',5'-Bis-[O-(1-oxooctyl)]-2'-O-methylcytidine



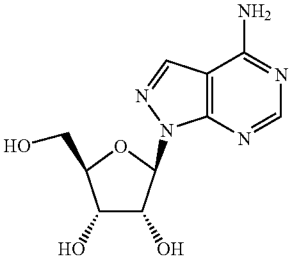
1,3-Dicyclohexylcarbodiimide (21.48 g, 104 mmol) was dissolved in anhydrous dichloromethane (100 mL). To the solution was added octanoic acid (5.49 mL, 34.5 mmol, made anhydrous by keeping over molecular sieves, 4 Å overnight at room temperature), and the resulting reaction mixture was stirred under argon atmosphere for 6 h. The white precipitate which formed was filtered, and the filtrate was concentrated under reduced pressure. The residue obtained was dissolved in anhydrous pyridine and added to N^4-(4,4'-dimethoxytrityl)-2'-O-methylcytidine (0.43 g, 0.77). DMAP (0.09 g, 0.77 mmol) was added and the resulting mixture was stirred at room temperature under argon atmosphere for 12 h. The solvent was removed under reduced pressure and the residue obtained was dissolved in ethyl acetate (100 mL). The organic phase was washed with

aqueous sodium bicarbonate (5%, 50 mL), dried over anhydrous $Na_2SO_4$and concentrated under reduced pressure. The residue was purified by flash silica gel column chromatography and eluted with 5% MeOH in dichloromethane. The product obtained was dissolved in a mixture of acetic acid: MeOH:H2O (20 mL, 3:6:1). The resulting mixture was heated at 50° C. for 24 h. The solvent was removed under reduced pressure. The residue obtained was purified by flash silica gel column chromatography and eluted with dichloromethane containing 0 to 5% of MeOH to give the title compound (0.22.g).

^{1}H NMR (200 MHz, DMSO-d_6) δ 0.83 (m, 611), 1.23 (br s, 16H), 1.51 (m, 4H), 2.33 (m, 4H), 3.26 (s, 31), 4.06 (t, J=5.2 Hz, 1H), 4.21 (m, 31), 5.11 (t, J=5.2 Hz, 1H), 5.75 (d, J3=7.4 Hz, 1H), 5.84 (d, J3=4.8 Hz, 1H), 7.26 (br s, 2H), 7.61 (d, J=7.4 Hz, 1H). MS (ES): m/z 510.3 $[M+H]^+$; HRMS (FAB) Calcd for $C_{26}H_{44}N_3O_7$: 510.3179; found 510.3170.

EXAMPLE 121

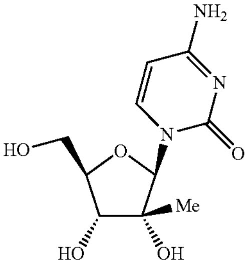
4-Amino-1-(β-D-ribofuranosyl)-1H-pyrazolo[3,4-d]pyrimidine



This compound was prepared following procedures described in *Nucleic Acids Res.*, 11: 871–872 (1983).

EXAMPLE 122

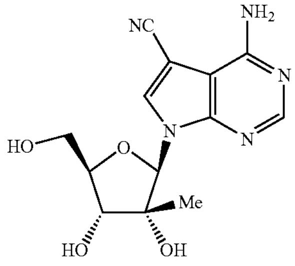
2' C-Methyl-cytidine



This compound was prepared following procedures described in L. Beigelman et al., *Carbohyd. Res.* 166: 219–232 (1987) or X-Q Tang, et al., *J. Org. Chem.* 64: 747–754 (1999).

EXAMPLE 123

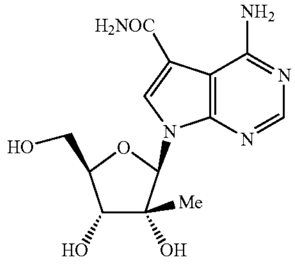
4-Amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile



This compound was prepared following procedures described by Y. Murai et al. in *Heterocycles* 33: 391–404 (1992).

EXAMPLE 124

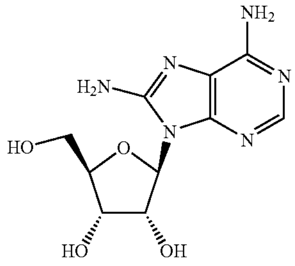
4-Amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyridine-5-carboxamide



This compound was prepared following procedures described by Y. Murai et al. in *Heterocycles* 33: 391–404 (1992).

EXAMPLE 125

8-Aminoadenosine



This compound was prepared following the procedure described in M. Ikehara and S. Yamada, *Chem. Pharm. Bull.*, 19: 104 (1971).

EXAMPLE 126

Mass Spectral Characterization of Nucleoside 5'-Triphosphates

Mass spectra of nucleoside 5'-triphosphates were determined as described in Example 87. Listed in the following table are the calculated and experimental masses for the nucleoside 5'-triphosphates prepared according to the procedures of Example 86. The example numbers correspond to the parent nucleoside of the nucleoside 5'-triphosphate.

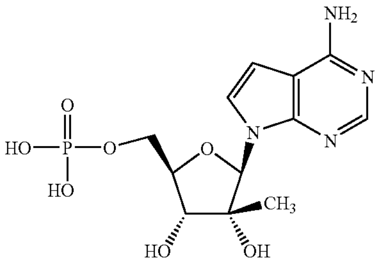
Example	Calculated	Found
1	507.0	506.9
2	525.0	524.9
5	537.0	537.0
6	539.0	539.0
7	565.0	565.0
8	547.0	546.9
9	550.0	550.0
10	506.0	505.9
11	536.0	535.9
12	536.0	536.0
13	561.0	560.9
14	550.0	550.0
15	524.0	524.0
16	522.0	521.9
17	547.0	546.9
18	536.0	536.0
20	531.0	530.9
21	522.0	522.0
22	536.0	536.0
23	506.0	506.1
24	524.0	524.0
25	508.0	508.0
26	508.0	508.0
27	552.0	552.0
28	506.0	506.0
29	579.0	578.9
30	582.0	582.0
31	568.0	567.9
32	554.0	553.9
33	540.0	539.9
34	554.0	553.9
35	568.0	567.9
36	541.0	541.0
37	565.0	564.9
38	542.0	541.9
39	554.0	553.9
41	481.0	481.0
42	467.0	467.0
43	485.0	484.8
46	482.0	482.0
47	486.0	485.8
48	482.0	482.0
49	554.0	554.0
51	468.0	468.1
52	521.0	521.0
53	491.0	491.2
55	584.9	585.1
56	521.0	521.2
58	506.0	506.0
61	520.0	519.9
62	520.0	520.0
63	547.0	547.0
64	533.0	533.0
65	549.0	549.0
67	551.0	551.0
68	515.0	514.9
69	520.0	520.1
71	490.0	489.9
89	523.0	522.9
90	521.0	520.9
91	645.1	645.0
94	524.0	523.9
95	522.0	521.8
98	536.0	535.9
99	520.0	520.0
102	613.1	613.0
103	498.0	497.9
104	481.0	481.0
105	536.0	536.2
106	509.0	508.9
108	505.0	505.0
112	506.0	506.1

-continued

Example	Calculated	Found
113	490.0	490.0
117	534.0	534.0
118	536.0	536.0

EXAMPLE 127

[4-Amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]-pyrimidine]-5'-monophosphate

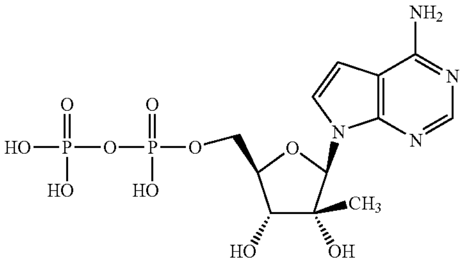


To the compound from Step F of Example 62 (14 mg, 0.05 mmol) (dried by coevaporation with pyridine and several times with toluene) was added trimethyl phosphate (0.5 mL). The mixture was stirred- overnight in a sealed container. It was then cooled to 0° C. and phosphorous oxychloride (0.0070 mL, 0.075 mmol) was added via a syringe. The mixture was stirred for 3 h at 0° C., then the reaction was quenched by addition of tetraethylammonium bicarbonate (TEAB) (1M) (0.5 mL) and water (5 mL). The reaction mixture was purified and analyzed according to the procedure described in Example 87.

Electron spray mass spectrum (ES-MS): Found: 359.2 (M–H$^+$), calc. for $C_{12}H_{17}N_4O_7P-H^+$: 359.1.

EXAMPLE 128

[4-Amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]-pyrimidine]-5'-diphosphate

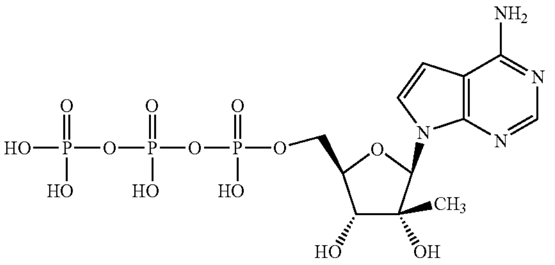


To the compound from Step F of Example 62 (56 mg, 0.20 mmol) (dried by coevaporation with pyridine and several times with toluene) was added trimethyl phosphate (stored over sieves) (1.0 mL). The mixture was stirred overnight in a sealed container. It was then cooled to 0° C. and phosphorous oxychloride (0.023 mL, 0.25 mmol) was added via a syringe. The mixture was stirred for 2 h at 0° C., then tributylamine (0.238 mL, 1.00 mmol) and tributylammonium phosphate (generated from phosphoric acid and tributylamine in pyridine, followed by repeated azeotropic evaporation with pyridine and acetonitrile) (1.0 mmol in 3.30 mL acetonitrile) was added. The mixture was stirred for an additional 30 min at 0° C., the sealed vial was then opened and the reaction quenched by addition of TEAB (1M) (1.0 mL) and water (5 mL). The reaction mixture was purified and analyzed according to the procedure described in Example 87.

ES-MS: Found: 439.0 (M–H$^+$), calc. for $C_{12}H_{18}N_4O_{10}P_2-H^+$: 439.04.

EXAMPLE 129

[4-Amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]-pyrimidine]-5'-triphosphate

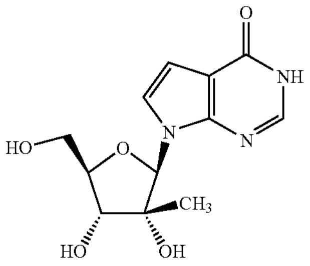


To the compound from Step F of Example 62 (20 mg, 0.07 mmol) (dried by coevaporation with pyridine and several times with toluene) was added trimethyl phosphate (stored over sieves) (0.4 mL). The mixture was stirred overnight in a sealed container. It was then cooled to 0° C. and phosphorous oxychloride (0.0070 mL, 0.075 mmol) was added via syringe. The mixture was stirred for 3 h at 0° C., then tributylamine (0.083 mL, 0.35 mmol), tributylammonium pyrophosphate (0.35 mmol, 127 mg) and acetonitrile (stored over sieves) (0.25 mL) were added. The mixture was stirred for an additional 30 min at 0° C., the sealed vial was then opened and the reaction quenched by addition of TEAB (1M) (0.5 mL) and water (5 mL). The reaction mixture was purified and analyzed according to the procedure described in Example 87.

ES-MS: Found: 519.0 (M–H$^+$), calc. for $C_{12}H_{19}N_4O_{13}P_3-H^+$: 519.01.

EXAMPLE 130

7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one

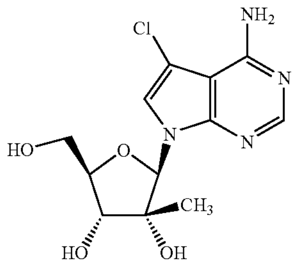


To the compound from Step E of Example 62 (59 mg, 0.18 mmol) was added aqueous sodium hydroxide (1M). The mixture was heated to reflux for 1 hr, cooled, neutralized with aqueous HCl (2M) and evaporated in vacuo. The residue was purified on silica gel using dichloromethane/methanol (4:1) as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (53 mg) as a colorless oil.

^{1}H NMR (CD_3CN): δ 0.70 (s, 3H), 3.34–4.15 (overlapping m, 7H), 6.16 (s, 1H), 6.57 (d, 3.6 Hz, 1H), 7.37 (d, 3.6 Hz, 1H), 8.83 (s, 1H).

EXAMPLE 131

4-Amino-5-chloro-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



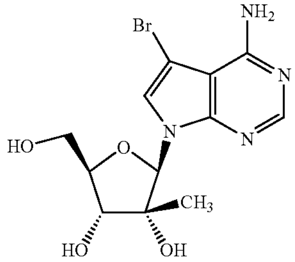
To a pre-cooled solution (0° C.) of the compound from Step F of Example 62 (140 mg, 0.50 mmol) in DMF (2.5 mL) was added N-chlorosuccinimide (0.075 g, 0.55 mmol) in DMF (0.5 mL) dropwise. The solution was stirred at rt for 1 h and the reaction quenched by addition of methanol (4 mL) and evaporated in vacuo. The crude product was purified on silica gel using methanol/dichloromethane (1:9) as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (55 mg) as a colorless solid.

^{1}H NMR (CD_3CN): δ 0.80 (s, 3H), 3.65–4.14 (overlapping m, 7H), 5.97 (s br, 2H), 6.17 (s, 1H), 7.51 (s, 1H), 8.16 (s, 1H).

ES-MS: Found: 315.0 (M+H$^+$), calc.for $C_{12}H_{15}ClN_4O_4$+H$^+$: 315.09.

EXAMPLE 132

4-Amino-5-bromo-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



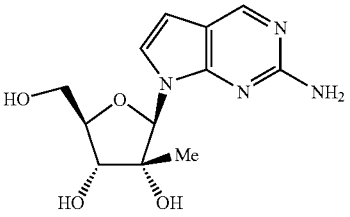
To a pre-cooled solution (0° C.) of the compound from Step F of Example 62 (28 mg, 0.10 mmol) in DMF (0.5 mL) was added N-bromosuccinimide (0.018 g, 0.10 mmol) in DMF (0.5 mL) dropwise. The solution was stirred at 0° C. for 20 min, then at rt for 10 min. The reaction was quenched by addition of methanol (4 mL) and evaporated in vacuo. The crude product was purified on silica gel using methanol/dichloromethane (1:9) as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (13.0 mg) as a colorless solid.

^{1}H NMR (CD_3CN): δ 0.69 (s, 3H), 3.46–4.00 (overlapping m, 7H), 5.83 (s br, 2H), 6.06 (s, 1H), 7.45 (s, 1H), 8.05 (s, 1H).

ES-MS: Found: 359.1 (M+H$^+$), calc. for $C_{12}H_{15}BrN_4O_4$+H$^+$: 359.04.

EXAMPLE 133

2-Amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

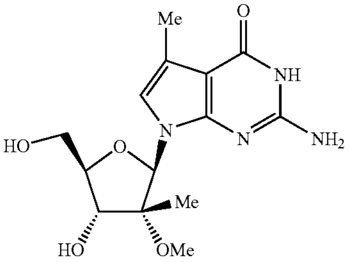


A mixture of 2-amino-4-chloro-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine (Example 118, Step B) (20 mg, 0.07 mmol) in EtOH (1.0 mL), pyridine (0.1 mL) and 10% Pd/C (6 mg) under H_2 (atmospheric pressure) was stirred overnight at room temperature. The mixture was filtered through a Celite pad which was thoroughly washed with EtOH. The combined filtrate was evaporated and purified on a silica gel column with CH_2Cl_2/MeOH, 20/1 and 10/1, as eluent to give the title compound as a white solid (16 mg).

^{1}H NMR (200 MHz, CD_3OD): δ 0.86 (s, 3H, 2'C-Me), 3.82 (dd, $J_{5',4'}$=3.6 Hz, $J_{5',5''}$=12.7 Hz, 1H, H-5'), 3.94–4.03 (m, 2H, H-5', H4'), 4.10 (d, $J_{3',4'}$=8.8 Hz, 1H, H-3'), 6.02 (s, 1H, H-1'), 6.41 (d, $J_{5,6}$=3.8 Hz, 1H, H-5), 7.39 (d, 1H, H-6), 8.43 (s, 1H, H-4). ES MS: 281.4 (MH$^+$).

EXAMPLE 134

2-Amino-5-methyl-7-(2-C,2-O-dimethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



Step A: 2-Amino-4-chloro-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C-methyl-β-D-ribofuranosy]-5-methyl-7H-pyrrolo[2,3-d]pyrimidine

To an ice-cold solution of the product from Step C of Example 62 (1.57 g, 3.16 mmol) in CH_2Cl_2 (50 mL) was added HBr (5.7 M in acetic acid; 3.3 mL) dropwise. The reaction mixture was stirred at 0° C. for 1 h and then at room temperature for 2 h, concentrated in vacuo and co-evaporated with toluene (2×20 mL). The resulting oil was dissolved in MeCN (20 mL) and added dropwise to a solution of the sodium salt of 2-amino-4-chloro-5-methyl-1H-pyrrolo[2,3-d]pyrimidine in acetonitrile [generated in situ from 2-amino-4-chloro-5-methyl-1H-pyrrolo[2,3-d]pyrimidine [for preparation, see *Liebigs Ann. Chem.* 1984: 708–721] (1.13 g, 6.2 mmol) in anhydrous acetonitrile (150 mL), and NaH (60% in mineral oil, 248 mg, 6.2 mmol), after

2 h of vigorous stirring at rt]. The combined mixture was stirred at rt for 24 h and then evaporated to dryness. The residue was suspended in water (100 mL) and extracted with EtOAc (300+150 mL). The combined extracts were washed with brine (100 mL), dried over Na_2SO_4, filtered and evaporated. The crude product was purified on a silica gel column (5×7 cm) using ethyl acetate/hexane (0 to 30% EtOAc in 5% step gradient) as the eluent. Fractions containing the product were combined and evaporated in vacuo to give the desired product (0.96 g) as a colorless foam.

Step B: 2-Amino-4-chloro-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C,2-O-dimethyl-β-D-ribofuranosyl]-5-methyl-7H-pyrrolo[2,3-d]pyrimidine

To an ice-cold mixture of the product from Step A (475 mg, 0.7 mmol) in THF (7 mL) was added NaH (60% in mineral oil, 29 mg) and stirred at 0° C. for 0.5 h. Then MeI (48 μL) was added and reaction mixture stirred at rt for 24 h. The reaction was quenched with MeOH and the mixture evaporated. The crude product was purified on a silica gel column (5×3.5 cm) using hexane/ethyl acetate (9/1, 7/1, 5/1 and 3/1) as eluent. Fractions containing the product were combined and evaporated to give the desired compound (200 mg) as a colorless foam.

Step C: 2-Amino-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C,2-O-dimethyl-β-D-ribofuranosyl]-5-methyl-7H-pyrrolo[2,3-d]pyrimidine-4(3H)-one

A mixture of the product from Step B (200 mg, 0.3 mmol) in 1,4-dioxane (15 mL) and aqueous NaOH (2N, 15 mL) in a pressure bottle was heated overnight at 135° C. The mixture was then cooled to 0° C., neutralized with 2N aqueous HCl and evaporated to dryness. The crude product was suspended in MeOH, filtered, and the solid thoroughly washed with MeOH. The combined filtrate was concentrated, and the residue purified on a silica gel column (5×5 cm) using CH_2Cl_2/MeOH (40/1, 30/1 and 20/1) as eluent to give the desired compound (150 mg) as a colorless foam.

Step D: 2-Amino-5-methyl-7-(2-C,2-O-dimethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one

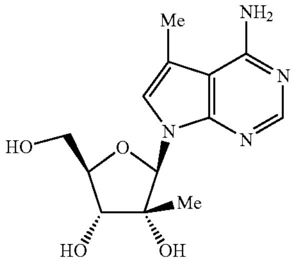
A mixture of the product from Step C (64 mg, 0.1 mmol) in MeOH (5 mL) and Et_3N (0.2 mL) and 10% Pd/C (24 mg) was hydrogenated on a Parr hydrogenator at 50 psi at r.t. for 1.5 days, then filtered through a Celite pad which was thoroughly washed with MeOH. The combined filtrate was evaporated and the residue purified on a silica gel column (3×4 cm) with CH_2Cl_2/MeOH (30/1, 20/1) as eluent to yield 2-amino-5-methyl-7-(5-O-benzyl-2-C,2-O-dimethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one. The compound (37 mg) was further hydrogenated in EtOH (2 mL) with 10% Pd/C and under atmospheric pressure of hydrogen. After stirring 2 days at r.t., the reaction mixture was filtered through Celite, the filtrate evaporated and the crude product purified on a silica gel column (1×7 cm) with CH_2Cl_2/MeOH (30/1, 20/1 and 10/1) as eluent to yield the title compound (12 mg) after freeze-drying.

^1H NMR (200 MHz, CD_3OD): δ 0.81 (s, 3H, 2'C-Me), 2.16 (d, $J_{H\text{-}6,C5\text{-}Me}$=1.3 Hz, 3H, C5-Me), 3.41 (s, 3H, 2'-OMe), 3.67 (dd, $J_{5'4'}$=3.4 Hz, $J_{5',5''}$=12.6 Hz, 1H, H-5'), 3.81–3.91 (m, 3H, H-5'', H-4', H-3'), 6.10 (s, 1H, H-1'), 6.66 (d, 1H, H-6).

ES MS: 323.3 $(M-H)^+$.

EXAMPLE 135

4-Amino-5-methyl-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



Step A: 4-Chloro-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C-methyl-β-D-ribofuranosyl]-5-methyl-7H-pyrrolo[2,3-d]pyrimidine

To an ice-cold solution of the product from Step C of Example 62 (1.06 g, 2.1 mmol) in CH_2Cl_2 (30 mL) was added HBr (5.7 M in acetic acid; 2.2 mL) dropwise. The reaction mixture was stirred at 0° C. for 1 h and then at room temperature for 2 h, concentrated in vacuo and co-evaporated with toluene (2×15 mL). The resulting oil was dissolved in MeCN (10 mL) and added dropwise into a solution of the sodium salt of 4-chloro-5-methyl-1H-pyrrolo[2,3-d]pyrimidine in acetonitrile [generated in situ from 4-chloro-5-methyl-1H-pyrrolo[2,3-d]pyrimidine [for preparation, see *J. Med. Chem.* 33: 1984 (1990)] (0.62 g, 3.7 mmol) in anhydrous acetonitrile (70 mL), and NaH (60% in mineral oil, 148 mg, 3.7 mmol), after 2 h of vigorous stirring at rt]. The combined mixture was stirred at rt for 24 h and then evaporated to dryness. The residue was suspended in water (100 mL) and extracted with EtOAc (250+100 mL). The combined extracts were washed with brine (50 mL), dried over Na_2SO_4, filtered and evaporated. The crude product was purified on a silica gel column (5×5 cm) using hexane/ethyl acetate (9/1, 5/1, 3/1) gradient as the eluent. Fractions containing the product were combined and evaporated in vacuo to give the desired product (0.87 g) as a colorless foam.

Step B: 4-Chloro-5-methyl-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

To a solution of the compound from Step A (0.87 g, 0.9 mmol) in dichloromethane (30 mL) at −78° C. was added boron trichloride (1M in dichloromethane, 9.0 mL, 9.0 mmol) dropwise. The mixture was stirred at −78° C. for 2.5 h, then at −30° C. to −20° C. for 3 h. The reaction was quenched by addition of methanol/dichloromethane (1:1) (9 mL) and the resulting mixture stirred at −15° C. for 30 min., then neutralized with aqueous ammonia at 0° C. and stirred at rt for 15 min. The solid was filtered and washed with CH_2Cl_2/MeOH (1/1, 50 mL). The combined filtrate was evaporated, and the residue was purified on a silica gel column (5×5 cm) using CH_2Cl_2 and CH_2Cl_2/MeOH (40/1 and 30/1) gradient as the eluent to furnish the desired compound (0.22 g) as a colorless foam.

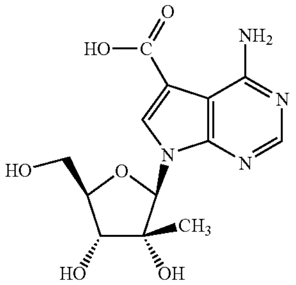
Step C: 4-Amino-5-methyl-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

To the compound from Step B (0.2 g, 0.64 mmol) was added methanolic ammonia (saturated at 0° C.; 40 mL). The mixture was heated in a stainless steel autoclave at 100° C. for 14 h, then cooled and evaporated in vacuo. The crude mixture was purified on a silica gel column (5×5 cm) with CH_2Cl_2/MeOH (50/1, 30/1, 20/1) gradient as eluent to give the title compound as a white solid (0.12 g).

^{1}H NMR (DMSO-d_6): δ 0.60 (s, 3H, 2'C-Me), 2.26 (s, 3H, 5C-Me), 3.52–3.61 (m, 1H, H-5'), 3.70–3.88 (m, 3H, H-5", H4', H-3'), 5.00 (s, 1H, 2'-OH), 4.91–4.99 (m, 3H, 2'-OH, 3'-OH, 5'-OH), 6.04 (s, 1H, H-1'), 6.48 (br s, 2H, NH2), 7.12 (s, 1H, H-6), 7.94 (s, 1H, H-2). ES MS: 295.2 (MH$^+$).

EXAMPLE 136

4-Amino-7-(2-C-methyl-β-D-ribofiuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carboxylic Acid

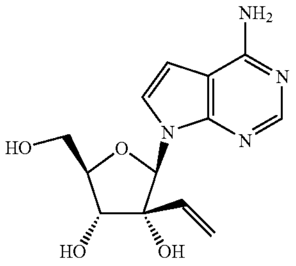


The compound of Example 123 (0.035 g, 0.11 mmol) was dissolved in a mixture of aqueous ammonia (4 mL, 30 wt %) and saturated methanolic ammonia (2 mL), and a solution of H_2O_2 in water (2 mL, 35 wt %) was added. The reaction mixture was stirred at room temperature for 18 h. Solvent was removed under reduced pressure, and the residue obtained was purified by HPLC on a reverse phase column (Altech Altima C-18, 10×299 mm, A=water, B=acetonitrile, 10 to 60% B in 50 min, flow 2 mL/min) to yield the title compound (0.015 g, 41%) as a white solid.

^{1}H NMR (CD_3OD): δ 0.85 (s, 3H, Me), 3.61 (m, 1H), 3.82 (m, 1H) 3.99–4.86 (m, 2H), 6.26 (s, 1H), 8.10 (s, 2H) 8.22(s, 1H); ^{13}C NMR (CD_3OD): 20.13, 61.37, 73.79, 80.42, 84.01, 93.00, 102.66, 112.07, 130.07, 151.40, 152.74, 159.12, 169.30. HRMS (FAB) Calcd for $C_{13}H_{17}N_4O_6+$ 325.1148, found 325.1143.

EXAMPLE 137

4-Amino-7-(2-C-vinyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



Step A: 3,5-Bis-O-(2,4-dichlorophenylmethyl)-2-C-vinyl-1-O-methyl-α-D-ribofuranose

Cerium chloride heptahydrate (50 g, 134.2 mmol) was finely crushed in a pre-heated mortar and transferred to a round-bottom flask equipped with a mechanical stirrer. The flask was heated under high vacuum overnight at 160° C. The vacuum was released under argon and the flask was cooled to room temperature. Anhydrous THF (300 mL) was cannulated into the flask. The resulting suspension was stirred at room temperature for 4 h and then cooled to −78° C. Vinylmagnesium bromide (1M in THF, 120 mL, 120 mmol) was added and stirring continued at −78° C. for 2 h. To this suspension was added a solution of 3,5-bis-O-(2,4-dichlorophenylmethyl)-1-O-methyl-α-D-erythro-pentofuranose-2-ulose (14 g, 30 mmol) [from Example 2, Step B] in anhydrous THF (100 mL), dropwise with constant stirring. The reaction was stirred at −78° C. for 4 h. The reaction was quenched with saturated ammonium chloride solution and allowed to come to room temperature. The mixture was filtered through a celite pad and the residue washed with Et_2O (2×500 mL). The organic layer was separated and the aqueous layer extracted with Et_2O (2×200 mL). The combined organic layers were dried over anhydrous Na_2SO_4 and concentrated to a viscous yellow oil. The oil was purified by flash chromatography (SiO_2, 10% EtOAc in hexanes). The title compound (6.7 g, 13.2 mmol) was obtained as a pale yellow oil.

Step B: 4-Chloro-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C-vinyl-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine

To a solution of the compound from Step A (6.4 g, 12.6 mmol) in anhydrous dichloromethane (150 mL) at −20° C. was added HBr (30% solution in AcOH, 20 mL, 75.6 mmol) dropwise. The resulting solution was stirred between −10° C. and 0° C. for 4 h, evaporated in vacuo and co-evaporated with anhydrous toluene (3×40 mL). The oily residue was dissolved in anhydrous acetonitrile (100 mL) and added to a solution of the sodium salt of 4-chloro-1H-pyrrolo[2,3-d]pyrimidine (5.8 g, 37.8 mmol) in acetonitrile (generated in situ as described in Example 62) at −20° C. The resulting mixture was allowed to come to room temperature and stirred at room temperature for 24 h. The mixture was then evaporated to dryness, taken up in water and extracted with EtOAc (2×300 mL). The combined extracts were dried over Na_2SO_4, filtered and evaporated. The crude mixture was purified by flash chromatography (SiO_2, 10% EtOAc in hexanes) and the title compound (1.75 g) isolated as a white foam.

Step, C: 4-Amino-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C-vinyl-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine

The compound from Step B (80, mg) was dissolved in the minimum amount of 1,4-dioxane and placed in a stainless steel bomb. The bomb was cooled to −78° C. and liquid ammonia was added. The bomb was sealed and heated at 90° C. for 24 h. The ammonia was allowed to evaporate and the residue concentrated to a white solid which was used in the next step without further purification.

Step D: 4-Amino-7-(2-C-vinyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyridine

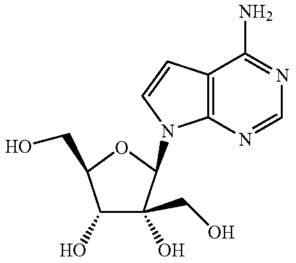
To a solution of the compound from Step C (60 mg) in dichloromethane at −78° C. was added boron trichloride (1M in dichloromethane) dropwise. The mixture was stirred at −78° C. for 2.5 h, then at −30° C. to −20° C. for 3 h. The reaction was quenched by addition of methanol/dichloromethane (1:1) and the resulting mixture stirred at −15° C. for 0.5 h, then neutralized with aqueous ammonia at 0° C. and stirred at room temperature for 15 min. The solid was filtered and washed with methanol/dichloromethane (1:1). The combined filtrate was evaporated and the residue purified by flash chromatography (SiO_2, 10% methanol in EtOAc containing 0.1% triethylamine). The fractions containing the product were evaporated to give the title compound as a white solid (10 mg).

^{1}H NMR (DMSO-d6): δ 3.6 (m, 1H, H-5'), 3.8 (m, 1H, H-5"), 3.9 (m d, 1-H, H4'), 4.3 (t, 1H, H-3'), 4.8–5.3(m, 6H, CH═CH$_2$, 2'-OH, 3'-OH, 5'-OH) 6.12 (s, 1H, H-1'), 6.59 (d, 1H, H-5), 7.1 (br s, 1H, NH2), 7.43 (d, 1H, H-6), 8.01 (s, 1H, H-2).

ES-MS: Found: 291.1 (M−H$^-$); calc. for $C_{13}H_{16}N_4O_4-H^-$: 291.2.

EXAMPLE 138

4-Amino-7-(2-C-hydroxymethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



Step A: 4-Chloro-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C-hydroxymethyl-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine

To a solution of the compound from Example 137, Step B (300 mg, 0.48 mmol) in 1,4-dioxane (5 mL) were added N-methylmorpholine-N-oxide (300 mg, 2.56 mmol) and osmium tetroxide (4% solution in water, 0.3 mL). The mixture was stirred in the dark for 14 h. The precipitate was removed by filtration through a celite plug, diluted with water (3×), and extracted with EtOAc. The EtOAc layer was dried over Na_2SO_4 and concentrated in vacuo. The oily residue was taken up in dichloromethane (5 mL) and stirred over $NaIO_4$ on silica gel (3 g, 10% $NaIO_4$) for 12 h. The silica gel was removed by filtration and the residue was evaporated and taken up in absolute ethanol (5 mL). The solution was cooled in an ice bath and sodium borohydride (300 mg, 8 mmol) was added in small portions. The resulting mixture was stirred at room temperature for 4 h and then diluted with EtOAc. The organic layer was washed with water (2×20 mL), brine (20 mL) and dried over Na_2SO_4. The solvent was evaporated and the residue purified by flash chromatography (SiO_2, 2:1 hexanes/EtOAc) to give the title compound (160 mg, 0.25 mmol) as white flakes.

Step B: 4-Amino-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C-hydroxymethyl-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine

The compound from Step A (150 mg, 0.23 mmol) was dissolved in the minimum amount of 1,4-dioxane (10 mL) and placed in a stainless steel bomb. The bomb was cooled to −78° C. and liquid ammonia was added. The bomb was sealed and heated at 90° C. for 24 h. The ammonia was allowed to evaporate and the residue concentrated to a white solid which was used in the next step without further purification.

Step C: 4-Amino-7-(2-C-hydroxymethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

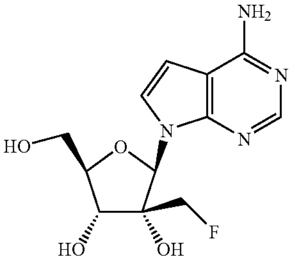
The compound from Step B (120 mg, 0.2 mmol) was dissolved in 1:1 methanol/dichloromethane, 10% Pd-C was added, and the suspension stirred under an H_2 atmosphere for 12 h. The catalyst was removed by filtration through a celite pad and washed with copious amounts of methanol. The combined filtrate was evaporated in vacuo and the residue was purified by flash chromatography (SiO_2, 10% methanol in EtOAc containing 0.1% triethylamine) to give the title compound (50 mg) as a white powder.

^1H NMR (CD_3OD): δ 3.12 (d, 1H, CH_2'), 3.33 (d, 1H, CH_2''), 3.82 (m, 1H, H-5'), 3.99–4.1(m, 2H, H-4', H-5''), 4.3 (d, 1H, H-3'), 6.2 (s, 1H, H-1'), 6.58 (d, 1H, H-5), 7.45 (d, 1H, H-6), 8.05 (s, 1H, H-2).

LC-MS: Found: 297.2 (M+H$^+$); calc. for $C_{12}H_{16}N_4O_5+H^+$: 297.3.

EXAMPLE 139

4-Amino-7-(2-C-fluoromethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



Step A: 4-Chloro-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C-fluoromethyl-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine

To a solution of the compound from Example 138, Step A (63 mg, 0.1 mmol) in anhydrous dichloromethane (5 mL) under argon, were added 4-dimethylaminopyridine (DMAP) (2 mg, 0.015 mmol) and triethylamine (62 μL, 0.45 mmol). The solution was cooled in an ice bath and p-toluenesulfonyl chloride (30 mg, 0.15 mmol) was added. The reaction was stirred at room temperature overnight, washed with $NaHCO_3$ (2×10 mL), water (10 mL), brine (10 mL), dried over Na_2SO_4 and concentrated to a pink solid in vacuo. The solid was dissolved in anhydrous THF (5 mL) and cooled in an icebath. Tetrabutylammonium fluoride (1M solution in THF, 1 mL, 1 mmol) was added and the mixture stirred at room temperature for 4 h. The solvent was removed in vacuo, the residue taken up in dichloromethane, and washed with $NaHCO_3$ (2×10 mL), water (10 mL) and brine (10 mL). The dichloromethane layer was dried over anhydrous Na_2SO_4, concentrated in vacuo, and purified by flash chromatography (SiO_2, 2:1 hexanes/EtOAc) to afford the title compound (20 mg) as a white solid.

Step B: 4-Amino-7-[3.5-bis-O-(2,4-dichlorophenylmethyl)-2-C-fluoromethyl-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine

The compound from Step A (18 mg, 0.03 mmol) was dissolved in the minimum amount of 1,4-dioxane and placed in a stainless steel bomb. The bomb was cooled to −78° C. and liquid ammonia was added. The bomb was sealed and heated at 90° C. for 24 h. The ammonia was allowed to evaporate and the residue concentrated to a white solid which was used in the next step without further purification.

Step C: 4-Amino-7-(2-C-fluoromethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

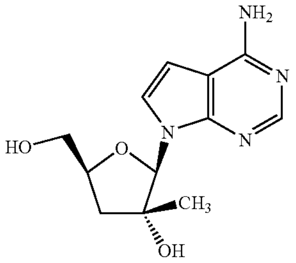
The compound from Step B (16 mg) was dissolved in 1:1 methanol/dichloromethane, 10% Pd—C was added, and the suspension stirred under an H_2 atmosphere for 12 h. The catalyst was removed by filtration through a celite pad and washed with copious amounts of methanol. The combined filtrate was evaporated in vacuo and the residue was purified by flash chromatography (SiO_2, 10% methanol in EtOAc containing 0.1% triethylamine) to give the title compound (8 mg) as a white powder.

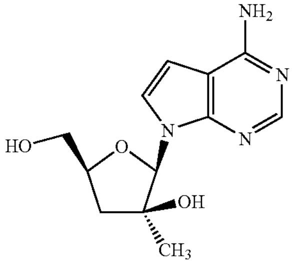
^1H NMR (DMSO-d_6): δ 3.6–3.7 (m, 1H, 1H-5'), 3.8–4.3 (m, 5H, H-5'', H-4', H-3', CH_2) 5.12 (t, 1H, 5'-OH), 5.35 (d, 1H, 3'-OH), 5.48 (s, 1H, 2'-OH), 6.21 (s, 1H, H-1'), 6.52 (d, 1H, H-5), 6.98 (br s, 2H, NH2), 7.44 (d, 1 H, H-6), 8.02 (s, 1H, H-2). ^{19}F NMR (DMSO-d_6): δ −230.2 (t).

ES-MS: Found: 299.1 (M+H$^+$), calc.for $C_{12}H_{15}FN_4O_4$+H$^+$: 299.27.

EXAMPLES 140 AND 141

4-Amino-7-(3-deoxy-2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine and 4-amino-7-(3-deoxy-2-C-methyl-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]-pyrimidine





Step A: 7-[2,5-Bis-O-(tert-butyldimethylsilyl)-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine and 7-[3,5-Bis-O-(tert-butyldimethylsilyl)-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine

To a stirred solution of tubercidin (5.0 g, 18.7 mmol) in a mixture of pyridine (7.5 mL) and DMF(18.5mL) was added silver nitrate (6.36 g, 38.8 mmol). This mixture was stirred at room temperature for 2 h. It was cooled in an ice bath and THF (37.4 mL) and tert-butyldimethylsilyl chloride (5.6 g, 37 mmol) was added and the mixture was stirred at room temperature for 2 h. The mixture was then filtered through a pad of celite and washed with THF. The filtrate and washings were diluted with ether containing a small amount of chloroform. The organic layer was washed successively with sodium bicarbonate and water (3×50 mL), dried over anhydrous sodium sulfate and concentrated. The pyridine was removed by coevaporation with toluene and the residue was purified by flash chromatography on silica gel using 5–7% MeOH in CH_2Cl_2 as the eluent; yield 3.0 g.

Step B: 7-[2,5-Bis-O-(tert-butyldimethylsilyl)-β-D-ribofuranosyl)]-4-[di-(4-methoxyphenyl)phenylmethyl]amino-7H-pyrrolo[2.3-d]pyrimidine and 7-[3,5-bis-O-(tert-butyldimethylsilyl)-β-D-ribofuranosyl]-4-[di-(4-methoxyphenyl)phenylmethyl]amino-7H-pyrrolo[2,3-d]pyrimidine

To a solution of mixture of the compounds from Step A (3.0 g, 6.0 mmol) in anhydrous pyridine (30 mL) was added 4,4'-dimethoxytrityl chloride (2.8 g, 8.2 mmol) and the reaction mixture was stirred at room temperature overnight. The mixture was then triturated with aqueous pyridine and extracted with ether. The organic layer was washed with water, dried over anhydrous sodium sulfate and concentrated to a yellow foam (5.6 g). The residue was purified by flash chromatography over silica gel using 20–25% EtOAc in hexanes as the eluent. The appropriate fractions were collected and concentrated to furnish 2',5'-O-bis-O-(tert-butyldimethylsilyl)- and 3',5'-bis-O-(tert-butyldimethylsilyl) protected nucleosides as colorless foams (2.2 g and 1.0 g, respectively).

Step C: 7-[2,5-Bis-O-(tert-butyldimethylsilyl)-3-O-tosyl-β-D-ribofuranosyl)]-4-[di-(4methoxyphenyl)phenylmethyl]amino-7H-pyrrolo[2,3-d]pyrimidine

To an ice-cooled solution of 2',5'-bis-O-(tert-butyldimethylsilyl)-protected nucleoside from Step B (2.0 g, 2.5 mmol) in pyridine (22 mL) was added p-toluenesulfonyl chloride (1.9 g, 9.8 mmol). The reaction mixture was stirred at room temperature for four days. It was then triturated with aqueous pyridine (50%, 10 mL) and extracted with ether (3×50 mL) containing a small amount of CH_2Cl_2 (10 mL). The organic layer was washed with sodium bicarbonate and water (3×30 mL). The organic layer was dried over anhydrous Na_2SO_4 and concentrated. Pyridine was removed by co-evaporation with toluene (3×25 mL). The residual oil was filtered through a pad of silica gel using hexane:ethyl acetate (70:30) as eluent; yield 1.4 g.

Step D: 4-[di-(4-methoxyphenyl)phenylmethyl]amino-7-[3-O-tosyl-β-D-ribofuranosyl-7H-pyrrolo[2,3-d]pyrimidine

A solution of the compound from Step C (1.0 g, 1.1 mmol) and THF (10 mL) was stirred with tetrabutylammonium fluoride (1M solution in THF, 2.5 mL) for 0.5 h. The mixture was cooled and diluted with ether (50 mL). The solution was washed with water (3×50 mL), dried over anhydrous Na_2SO_4, and concentrated to an oil. The residue was purified by passing through a pad of silica gel using hexane: ethyl acetate (1:1) as eluent; yield 780 mg.

Step E: 4-Amino-7-(3-deoxy-2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]-pyrimidine and 4-amino-7-(3-deoxy-2-C-methyl-β-D-arabinofuranosyl)-7H-pyrrolo-[2,3-d]pyrimidine

A solution of CH_3MgI (3.0 M solution in ether, 3.0 mL) in anhydrous toluene (3.75 mL) was cooled in an ice bath. To this was added a solution of the compound from Step D (500 mg, 0.8 mmol) in anhydrous toluene (3.7 mL). The resulting mixture was stirred at room temperature for 3.5 h. It was cooled and treated with aqueous NH_4Cl solution and extracted with ether (50 mL containing 10 mL of CH_2Cl_2). The organic layer was separated and washed with brine (2×30 mL) and water (2×25 mL), dried over anhydrous Na_2SO_4 and concentrated to an oil which was purified by flash chromatography on silica gel using 4% MeOH in CH_2Cl_2 to furnish the 2-C-α-methyl compound (149 mg) and the 2-C-β-methyl compound (34 mg). These derivatives were separately treated with 80% acetic acid and the reaction mixture stirred at room temperature for 2.5 h. The acetic acid was removed by repeated co-evaporation with ethanol and toluene. The residue was partitioned between chloroform and water. The aqueous layer was washed with chloroform and concentrated. The evaporated residue was purified on silica gel using 5–10% MeOH in CH_2Cl_2 as the eluent to furnish the desired compounds as white solids.

4-Amino-7-(3-deoxy-2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine (9.0 mg)

^{1}H NMR (DMSO-d_6): δ 0.74 (s, 3H, CH_3), 1.77 (dd, 1H, H-3'), 2.08 (t, 1H, H-3"), 3.59 (m, 1H, H-5'), 3.73 (m, 1H, H-5"), 4.15 (m, 1H, H-4'), 5.02 (t, 1H, OH-5'), 5.33 (s, 1H, OH-2'), 6.00 (s, 1H, H-1'), 6.54 (d, 1H, H-7), 6.95 (br s, 2H, NH_2), 7.47 (d, 1H, H-8), 8.00 (s, 1H, H-2); ES-MS: 263.1 [M–H].

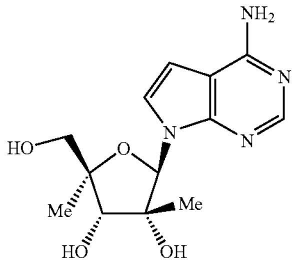
4-Amino-7-(3-deoxy-2-C-methyl-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine (15 mg):

^{1}H NMR (DMSO-d_6): δ 1.23 (s, 3H, CH_3), 2.08 (ddd, 2H, H-3 and 3"), 3.57 (m, 2H, H-5' and 5"), 4.06 (m, 1H, H4), 5.10 (s, 1H, OH-2'), 5.24 (t, 1H, OH-5'), 6.01 (s, 1H, H-1'), 6.49 (d, 1H, H-7), 6.89 (br s, 2H, NH_2), 7.35 (d, 1H, H-8), 8.01 (s,1H,H-2).

ES-MS: 265.2[M+H].

EXAMPLE 142

4-Amino-7-(2,4-C-dimethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



Step A: 5-Deoxy-1,2-O-isopropylidene-D-xylofuranose

1,2-O-Isopropylidene-D-xylofuranose (38.4 g, 0.2 mol), 4-dimethylaminopyridine (5 g), triethylamine (55.7 mL, 0.4 mol) were dissolved in dichloromethane (300 mL). p-Toluenesulfonyl chloride (38.13 g, 0.2 mol) was added and the reaction mixture was stirred at room temperature for 2 h. The reaction mixture was then poured into saturated aqueous sodium bicarbonate (500 mL) and the two layers were separated. The organic layer was washed with aqueous citric acid solution (20%, 200 mL), dried (Na_2SO_4) and evaporated to give a solid (70.0 g). The solid was dissolved in dry THF (300 mL) and $LiAlH_4$ (16.0 g, 0.42 mol) was added in portions over 30 min. The mixture was stirred at room temperature for 15 h. Ethyl acetate (100 mL) was added dropwise over 30 min and the mixture was filtered through a silica gel bed. The filtrate was concentrated and the resulting oil was chromatographed on silica gel (EtOAc/hexane 1/4) to afford the product as a solid (32.5 g).

Step B: 3,5-Bis-O-(2,4-dichlorophenylmethyl)-1-O-methyl-4-methyl-α-D-ribofuranose

Chromium oxide (50 g, 0.5 mol), acetic anhydride (50 mL, 0.53 mol) and pyridine (100 mL, 1.24 mol) were added to dichloromethane (1 L) in an ice water bath and the mixture was stirred for 15 min. 5-Deoxy-1,2-O-isopropylidene-D-xylofuranose (32 g, 0.18 mol) in dichloromethane (200 mL) was added, and the mixture was stirred at the same temperature for 30 min. The reaction solution was diluted with ethyl acetate (1 L) and filtered through a silica gel bed. The filtrate was concentrated to give a yellow oil. The oil was dissolved in 1,4-dioxane (1 L) and formaldehyde (37%, 200 mL). The solution was cooled to 0° C. and solid KOH (50 g) was added. The mixture was stirred at room temperature overnight and was then extracted with ethyl acetate (6×200 mL). After concentration, the residue was chromatographed on silica gel (EtOAc) to afford the product as an oil (1.5 g). The oil was dissolved in 1-methyl-2-pyrrolidinone (20 mL) and 2,4-dichlorophenylmethyl chloride (4 g, 20.5 mmol) and NaH (60%, 0.8 g) were added. The mixture was stirred overnight and diluted with toluene (100 mL). The mixture was then washed with saturated aqueous sodium bicarbonate (3×50 mL), dried (Na_2SO_4) and evaporated. The residue was dissolved in methanol (50 mL) and HCl in dioxane (4 M, 2 mL) was added. The solution was stirred overnight and evaporated. The residue was chromatographed on silica gel (EtOAc/hexane:1/4) to afford the desired product as an oil (2.01 g).

Step C: 3,5-Bis-O-(2,4-dichlorophenylmethyl)-2,4-di-C-methyl-1-O-methyl-α-D-ribofuranose

The product (2.0 g, 4.0 mmol) from Step B and Dess-Martin periodinane (2.0 g) in dichloromethane (30 mL) were stirred overnight at room temperature and was then concentrated under reduced pressure. The residue was triturated with ether ether (50 mL) and filtered. The filtrate was washed with a solution of $Na_2S_2O_3.5H_2O$(2.5 g) in saturated aqueous sodium bicarbonate solution (50 mL), dried ($MgSO_4$), filtered and evaporated. The residue was dissolved in anhydrous Et_2O (20 mL) and was added dropwise to a solution of MeMgBr in Et_2O (3 M, 10 mL) at −78° C. The reaction mixture was allowed to warm to −30° C. and stirred at −30° C. to −15° C. for 5 h, then poured into saturated aqueous ammonium chloride (50 mL). The two layers were separated and the organic layer was dried ($MgSO_4$), filtered and concentrated. The residue was chromatographed on silica gel (EtOAc/hexane: 1/9) to afford the title compound as a syrup (1.40 g).

Step D: 4-Chloro-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2,4-di-C-methyl-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine

To the compound from Step C (0.70 g, 1.3 mmol) was added HBr (5.7 M in acetic acid, 2 mL). The resulting solution was stirred at room temperature for 1 h, evaporated in vacuo and co-evaporated with anhydrous toluene (3×10 mL). 4-Chloro-1H-pyrrolo[2,3-d]pyrimidine (0.5 g, 3.3 mmol) and powdered KOH (85%, 150 mg, 2.3 mmol) were stirred in 1-methyl-2-pyrrolidinone (5 mL) for 30 min and the mixture was co-evaporated with toluene (10 mL). The resulting solution was poured into the above bromo sugar residue and the mixture was stirred overnight. The mixture was diluted with toluene (50 mL), washed with water (3×50 mL) and concentrated under reduced pressure. The residue was chromatographed on silica gel eluting with (EtOAc/Hexane 15/85) to afford a solid (270 mg).

Step E: 4-Amino-7-(2,4-di-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

The compound from Step D (270 mg) was dissolved in dioxane (2 mL) and liquid ammonia (20 g) was added in a stainless steel autoclave. The mixture was heated at 100° C. for 15 h, then cooled and evaporated. The residue was chromatographed on silica gel (EtOAc) to afford a solid (200 mg). The solid (150 mg) and Pd/C (10% 150 mg) in methanol (20 mL) were shaken under H_2 (30 psi) for 3 h, filtered and evaporated. The residue was chromatographed on silica gel ($MeOH/CH_2Cl_2$: 1/9) to afford the desired product as a solid (35 mg).

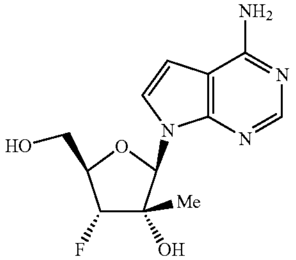
^1H NMR (DMSO-d_6): δ 0.65 (s, 3H), 1.18 (s, 3H), 3.43 (m, 2H), 4.06 (d, 1H, J 6.3 Hz), 4.87 (s, 1H), 5.26 (br, 1H), 5.08 (d, 1H, J 6.3 Hz), 5.25 (t, 1H, J 3.0 Hz), 6.17 (s, 1H), 6.54 (d, 1H, J 3.5 Hz), 6.97 (s, br, 2H), 7.54 (d, 1H, J 3.4 Hz), 8.02 (s, 1H).

^{13}C NMR (DMSO-d_6): δ 18.19, 21.32, 65.38, 73.00, 79.33, 84.80, 90.66, 99.09, 102.41, 121.90, 149.58, 151.48, 157.38.

LC-MS: Found: 295.1 ($M+H^+$); calculated for $C_{13}H_{18}N_4O_4+H^+$: 295.1

EXAMPLE 143

4-Amino-7-(3-deoxy-3-fluoro-2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



Step A: 3-Deoxy-3-fluoro-1-O-methyl-5-O-toluoyl-α-D-ribofuranose

1,2-O-Isopropylidene-D-xylofuranose (9.0 g, 50 mmol) and p-toluoyl chloride (7.0 mL, 50 mmol) in pyridine (50 mL) were stirred for 30 min. Water (10 mL) was added and the mixture was concentrated under reduced pressure. The residue was dissolved in toluene (500 mL) and the solution was washed with water (200 mL) and saturated aqueous sodium bicarbonate (200 mL). The two layers were separated and the organic layer was evaporated. The residue was dissolved in methanol (100 mL) and HCl in dioxane (4 M, 10 mL) was added. The mixture was stirred at room temperature overnight and was then evaporated under reduced pressure. The resulting oil was chromatographed on silica gel (EtOAc/hexane: 1/1) to afford an oil (10.1 g). The oil was dissolved in dichloromethane (100 mL) and diethylaminosulfur trifluoride (DAST) (5.7 mL) was added. The mixture was stirred overnight and was then poured into saturated aqueous sodium bicarbonate solution (100 mL). The mixture was extracted with toluene (2×50 mL) and the combined organic layers were concentrated. The residue was chromatographed on silica gel (EtOAc/hexane: 15/85) to afford the title compound as an oil (1.50 g).

Step B: 3-Deoxy-3-fluoro-2-C-methyl-1-O-methyl-5-O-toluoyl-α-D-ribofuranose

The product from Step A (1.0 g, 3.5 mmol) and Dess-Martin periodinane (2.5 g) in dichloromethane (20 mL) were stirred overnight at room temperature and was then concentrated under reduced pressure. The residue was triturated with diethyl ether (50 mL) and filtered. The filtrate was washed with a solution of $Na_2S_2O_3.5H_2O$ (12.5 g) in saturated aqueous sodium bicarbonate (100 mL), dried ($MgSO_4$), filtered and evaporated. The residue was dissolved in anhydrous THF (50 mL). $TiCl_4$ (3 mL) and methyl magnesium bromide in ethyl ether (3 M, 10 mL) were added at −78° C. and the mixture was stirred at −50 to −30° C. for 2 h. The mixture was poured into saturated aqueous sodium bicarbonate solution (100 mL) and filtered through Celite. The filtrate was extracted with toluene (100 mL) and evaporated. The residue was chromatographed on silica gel (EtOAc/hexane: 15/85) to afford the title compound as an oil (150 mg).

Step C: 4-Amino-7-(3-deoxy-3-fluoro-2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyriridine

The product from Step B (150 mg, 0.5 mmol) was dissolved in HBr (30%) in acetic acid (2 mL). After one hour, the mixture was evaporated under reduced pressure and co-evaporated with toluene (10 mL). 4-Chloro-1H-pyrrolo[2,3-d]pyrimidine (0.5 g, 3.3 mmol) and powdered KOH (85%, 150 mg, 2.3 mmol) were stirred in DMF (3 mL) for 30 min and the mixture was co-evaporated with toluene (2 mL). The resulting solution was poured into the above bromo sugar and the mixture was stirred overnight. The mixture was diluted with toluene (50 mL), washed with water (3×50 mL) and concentrated under reduced pressure. The residue was chromatographed on silica gel (EtOAc/hexane 15/85) to afford an oil (60 mg). The oil was dissolved in dioxane (2 mL) and liquid ammonia (20 g) was added in a stainless steel autoclave. The mixture was heated at 85° C. for 18 h, then cooled and evaporated. The residue was chromatographed on silica gel (methanol/dichloromethane: 1/9) to afford the title compound as a solid (29 mg).

^1H NMR (DMSO-d_6): δ 0.81 (s, 3H), 3.75 (m, 2H), 4.16 (m, 1H), 5.09 (dd, 1H, J 53.2, 7.8 Hz), 5.26 (br, 1H), 5.77 (s, 1H), 6.15 (d, 1H, J 2.9 Hz), 6.59 (d, 1H, J 3.4 Hz), 7.02 (s br, 2H), 7.39 (d, 1H, J 3.4 Hz), 8.06 (s, 1H).

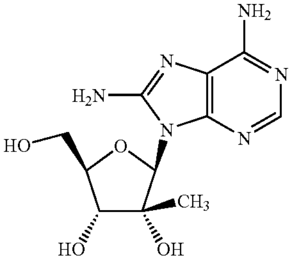
^{13}C NMR (DMSO-d_6): 19.40, 59.56, 77.24, 79.29, 90.15, 91.92, 99.88, 102.39, 121.17, 149.80, 151.77, 157.47.

^{19}F NMR (DMSO-d_6): δ 14.66 (m).

ES-MS: Found: 283.1 ($M+H^+$); calculated for $C_{12}H_{15}FN_4O_3+H^+$: 283.1.

EXAMPLE 144

8-Amino-2'-C-methyladenosine



Step A: 8-Bromo-2'-C-methyladenosine

To a solution of 2'-C-methyladenosine [for preparation, see *J. Med. Chem.* 41: 1708 (1998)] (138 mg, 0.5 mmol) in DMF (4 mL) was added N-bromosuccinimide (231 mg, 1.35 mmol). The solution was stirred protected from light at rt for 2 d and then evaporated in vacuo. The crude product was purified on a silica gel column (3×9 cm) using dichloromethane/methanol (25/1, 20/1 and 15/1) as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (38 mg) as a white solid.

Step B: 8-Amino-2'-C-methyladenosine

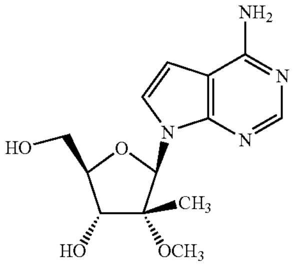
A solution of the compound from Step A (38 mg, 0.11 mmol) in liquid ammonia (10 mL) was heated in a stainless steel autoclave at 105° C. for 1 d, then cooled and evaporated. The residue was purified by HPLC [C-18 Phenomenex Luna (10μ; 250×21.2 mm) RP-column; solvents: (A) water, (13) acetonitrile; Linear gradient: 2–35% B in 76 min.] to yield the title compound (12 mg) as a white fluffy material after freeze-drying.

^1H NMR (DMSO-d_6): δ 0.70 (s, 3H, Me), 3.55–3.75 (m, 3H, H-5', H-5", H-4'), 4.03 (m, 1H, H-3'), 4.81 (s, 1H, 2'-OH), 5.10 (d, 1H, 3'-OH), 5.45 (t, 1H, 5'-OH), 5.86 (s, 1H, H-1'), 6.30, 6.39 (2s, 6H, 2 NH_2), 7.78 (s, 1H, H-2).

ES-MS: Found: 295.0 ($M-H^+$).

EXAMPLE 145

4-Amino-7-(2-C,2-O-dimethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



Step A: 4-chloro-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C,2-O-dimethyl-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine

To a pre-cooled (0° C.) solution of the compound from Example 62, Step D (618 mg, 1.0 mmol) in THF (8 mL) was added methyl iodide (709 mg, 5.0 mmol) and NaH (60% in mineral oil) (44 mg, 1.1 mmol). The resulting mixture was stirred overnight at rt and then poured into a stirred mixture of saturated aqueous ammonium chloride (50 mL) and dichloromethane (50 mL). The organic layer was washed with water (50 mL), dried ($MgSO_4$) and evaporated in vacuo. The resulting crude product was purified on silica gel using ethyl acetate/hexane as the eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (735 mg) as a colorless foam.

Step B: 4-amino-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C,2-O-dimethyl-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine

To the compound from Step A (735 mg, 1.16 mmol) was added methanolic ammonia (saturated at 0° C.) (20 mL). The mixture was heated in a stainless steel autoclave at 80° C. overnight, then cooled and the content evaporated in vacuo. The crude mixture was purified on silica gel using ethyl acetate/hexane as the eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (504 mg) as colorless foam.

Step C: 4-amino-7-(2-C,2-O-dimethyl-β-D-ribofiuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

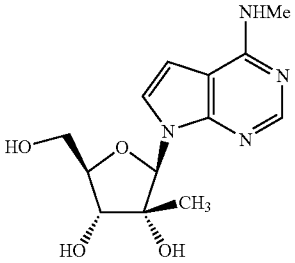
A mixture of the product from Step C (64 mg, 0.1 mmol), MeOH (5 mL), Et_3N (0.2 mL) and 10% Pd/C (61 mg) was hydrogenated on a Parr hydrogenator at 50 psi at room temperature overnight. The mixture was filtered throught celite, evaporated in vacuo and filtered through a pad of silica gel using 2% methanol in dichloromethane as eluent. The desired product was collected and evaporated in vacuo. The compound was redissolved in methanol (10 mL) and 10% Pd/C (61 mg) was added. The mixture was hydrogenated on a Parr hydrogenator at 55 psi at room temperature for two weeks. The mixture was filtered through celite, evaporated in vacuo and purified on silica gel using 10% methanol in dichloromethane as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (110 mg) as a colorless foam.

^1H NMR (DMSO-d_6): δ 0.68 (s, 3H,), 3.40 (s, 3H), 3.52–3.99 (overlapping m, 4H), 4.92 (d, 1H), 5.07 (t, 1H), 6.26 (s, 1H), 6.55 (d, 1H), 7.00s br, 2H), 7.46 (d, 1H), 8.05 (s, 1H).

LC-MS: Found: 293.1 (M−H^+); calc. for $C_{12}H_{16}N_4O_4$−H^+: 293.12.

EXAMPLE 146

4-Methylamino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



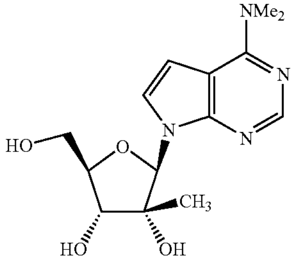
The compound from Step E of Example 62 (200 mg, 0.67 mmol) was added to methylamine (5 mL condensed in a small stainless steel autoclave) and warmed at 85° C. for 48 h, then cooled and evaporated in vacuo. The crude mixture was purified on a silica gel with ethanol as the eluent to give the title compound which separated as an amorphous solid after treatment with MeCN. The amorphous solid was dissolved in water and lyophilized to give a colorless powder (144 mg).

^1H NMR (DMSO-d_6): δ 0.63 (s, 3H, CH_3), 3.32 (s, 3H, N CH_3), 3.58–3.67 (m, 1H, H-5'), 3.79–3.39 (m, 3H, H-5", H4', H-3'), 5.03 (s, 1H, 2'-OH), 5.04–5.11 (1H,3'-OH, 1H, 5'-OH), 6.14 (s, 1H, H-1'), 6.58 (d, 1H, $J_{5,6}$=3.6 Hz, H-5), 7.46 (d, 1H, H-6), 7.70 (br s, 1H, NH), 8.14 (s, 1H, H-2).

LC-MS: Found: 295.1 (M−H^+); calc. for $C_{13}H_{18}N_4O_4$+H^+: 294.3.

EXAMPLE 147

4-Dimethylamino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



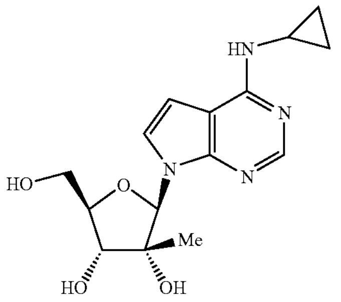
The compound from Step E of Example 62 (200 mg, 0.67 mmol) was added to dimethylamine (5 mL condensed in a small stainless steel autoclave) and warmed at 85° C. for 48 h, then cooled and evaporated in vacuo. The crude mixture was purified on a silica gel with ethanol as the eluent to give the title compound which separated as an amorphous solid after treatment with MeCN. The amorphous solid was dissolved in water and lyophilized to give a colorless powder (164 mg).

^1H NMR (DMSO-d_6): δ 0.64 (s, 3H, CH_3), 3.29 (s, 3H, N CH_3), 3.32 (s, 3H, N CH_3), 3.60–3.66 (m, 1H, H-5'), 3.77–3.97 (m, 3H, H-5", H-4', H-3'), 5.04 (s, 1H, 2'-OH), 5.06–5.11 (1H, 3'-OH, 1H, 5'-OH), 6.21 (s, 1H, H-1'), 6.69 (d, 1H, $J_{5,6}$=3.6 Hz, H-5), 7.55 (d, 1H, H-6), 8.13 (s, 1H, H-2).

LC-MS: Found: 309.3 (M−H^+); calc. for $C_{14}H_{20}N_4O_4$+H^+: 308.33.

EXAMPLE 148

4-Cyclopropylamino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



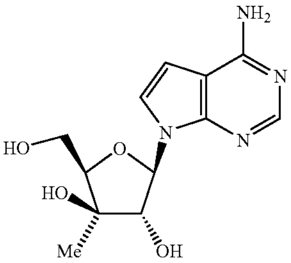
The compound from Step E of Example 62 (200 mg, 0.67 mmol) was added to cyclopropylamine (5 mL condensed in a small stainless steel autoclave) and warmed at 85° C. for 48 h, then cooled and evaporated in vacuo. The crude mixture was purified on a silica gel with ethanol as the eluent to give the title compound which separated as an amorphous solid after treatment with MeCN. The amorphous solid was dissolved in water and lyophilized to give a colorless powder (148 mg).

^{1}H NMR (DMSO-d_6): δ 0.51–0.58 (m, 2H), 0.64 (s, 3H, CH_3), 0.74–0.076 (m, 2H), 3.62–3.67 (m, 1H, H-5'), 3.79–3.82 (m, 3H, H-5"), 3.92–3.96 (m, H-4', H-3'), 5.03 (s, 1H, 2'-OH), 5.05–5.10 (1H, 3'-OH, 1H, 5'-OH), 6.15 (s, 1H, H-1'), 7.48 (d, 1H, $J_{5,6}$=3.6 Hz, H-5), 7.59 (d, 1H, H-6), 8.13 (s, 1H, H-2).

LC-MS: Found: 321.1 (M–H^+); calc. for $C_{15}H_{20}N_4O_4$+H^+: 320.3.

EXAMPLE 149

4-Amino-7-(3-C-methyl-β-D-xylofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



Step A: 7-[2,5-Bis-O-(tert-butyldimethylsilyl)-β-D-ribofuranosyl)]-4-[(4-methoxyphenyl)diphenylmethyl]amino-7H-pyrrolo[2,3-d]pyrimidine and 7-[3,5-bis-O-(tert-butyldimethylsilyl)-β-D-ribofuranosyl]-4-[(4-methoxyphenyl)diphenylmethyl]amino-7H-pyrrolo[2,3-d]pyrimidine

To a solution of mixture of the compounds from Step A of Examples 140 and 141 (0.32 g, 0.65 mmol) in anhydrous pyridine (6 mL) was added monomethoxytrityl chloride (0.30 g, 0.98 mmol) and the reaction mixture was stirred at room temperature overnight. The mixture was then concentrated and the residue was partitioned between CH_2Cl_2 (70 mL) and water (20 mL). The organic layer was washed with water and brine, dried (Na_2SO_4) and concentrated. The residue was purified on silica gel column using 5–13% EtOAc in hexanes as the eluent. The appropriate fractions were collected and concentrated to furnish 2',5'-bis-O-(tert-butyldimethylsilyl)- and 3',5'-bis-O-(tert-butyldimethylsilyl) protected nucleosides as colorless foams (343 mg and 84 mg, respectively).

Step B: 7-[2,5-Bis-O-(tert-butyldimethylsilyl)-β-D-erythro-pentofuranos-3-ulosyl]-4-[(4-methoxyphenyl)diphenylmethyl]amino-7H-pyrrolo[2,3-d]pyrimidine

To a well-stirred suspension of chromium trioxide (91 mg, 0.91 mmol) in CH_2Cl_2 (4 mL) at 0° C. were added pyridine (147 μL, 1.82 mmol) and then acetic anhydride (86 μL, 0.91 mmol). The mixture was stirred at room temperature for 0.5 h. Then the 2',5'-bis-O-(tert-butyldimethylsilyl) protected nucleoside from step A (343 mg 0.45 mmol) in CH_2Cl_2 (2.5 mL) was added and the mixture stirred at room temperature 2 h. The mixture was then poured into ice-cold EtOAc (10 mL) and filtered through a short silica gel column using EtOAc as the eluent. The filtrate was evaporated and the residue purified on a silica gel column with hexanes and hexanes/EtOAc (7/1) as the eluent to give the title compound (180 mg).

Step C: 7-[2,5-Bis-O-(tert-butyldimethylsilyl)-3-C-methyl-β-D-ribofuranosyl)-4-[(4-methoxyphenyl)diphenylmethyl]amino-7H-pyrrolo[2,3-d]pyrimidine and 7-[2,5-Bis-O-(tert-butyldimethylsilyl)-3-C-methyl-β-D-xylofuranosyl)-4-[(4-methoxyphenyl)diphenylmethyl]amino-7H-pyrrolo[2,3-d]pyrimidine

To a mixture of MeMgBr (3.0 M solution in ether; 0.17 mL, 0.5 mmol) in anhydrous hexanes (1.5 mL) at room temperature was added dropwise a solution of the compound from Step B (78 mg, 0.1 mmol) in anhydrous hexanes (0.5 mL). After 2 h stirring at room temperature, the reaction mixture was poured into ice-cold water (10 mL) and diluted with EtOAc (20 mL), then filtered through Celite which was then thoroughly washed with EtOAc. The layers were separated and the organic layer was washed with brine, dried (Na_2SO_4) and concentrated. The residue was purified on a silica gel column using 8 to 25% EtOAc in hexanes as eluent to give the 3-C-methyl xylo- (60 mg) and the 3-C-methyl ribo-isomer (20 mg).

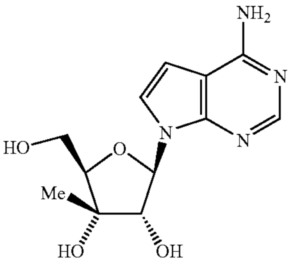
Step D: 4-Amino-7-(3-C-methyl-β-D-xylofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

To an ice-cold solution of 3-C-methyl-xylo isomer from Step C (60 mg, 0.08 mmol) in THF (2 mL) was added TBAF (1 M in THF; 0.32 ml, 0.32 mmol). The reaction mixture was stirred at room temperature for 5 h, then diluted with CH_2Cl_2 (50 mL), washed with water (3×15 mL), dried,and evaporated. The residue was dissolved in dioxane (0.3 mL) and 80% acetic acid (3 mL) was added. The reaction mixture was stirred at room temperature for 1 d and then evaporated. The residue was co-evaporated with dioxane, taken up in water (50 mL) and washed with CH_2Cl_2 (2×10 mL). The aqueous layer was concentrated and then freeze-dried. The residue was purified on silica gel column with CH_2Cl_2/MeOH (20/1 and 10/1) as the eluent to give the title compound as a white fluffy compound after freeze drying (10 mg).

^{1}H NMR (CD_3CN): δ 1.28 (s, 3H, CH_3), 3.56 (br s, 1H, OH), 3.78 (m, 3H, H-4', H-5', H-5"), 4.10 (br s, 1H, OH), 4.44 (d, 1H, $J_{2',1'}$=3.9 Hz, H-2'), 5.58 (d, 1H, H-1'), 5.85 (br s, 2H, NH_2), 6.15 (br s, 1H, OH), 6.48 (d, 1H, $J_{5,6}$=3.7 Hz, H-5), 7.23 (d, 1H, H-6), 8.11 (s, 1H, H-2). ES-MS: 281 $[MH]^+$.

EXAMPLE 150

4-Amino-7-(3-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

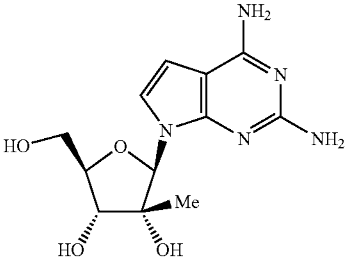


The ribo-isomer (20 mg) from Step C of Example 149 was deprotected using the procedure described in Step D of Example 32 to yield the title compound (4 mg).

^{1}H NMR (CD$_3$CN): δ 1.43 (s, 3H, CH$_3$), 3.28 (br s, 1H, OH), 3.58 (m, 2H, H-5', H-5"), 3.99 (m, 1H, H-4'), 4.10 (br s, 1H, OH), 4.62 (d, 1H, $J_{2',1'}$=8.1 Hz, H-2'), 5.69 (d, 1H, H-1'), 5.88 (br s, 3H, OH, NH$_2$), 6.45 (br s, 1H, OH), 6.51 (d, 1H, $J_{5,6}$=3.7 Hz, H-5), 7.19 (d, 1H, H-6), 8.12 (s, 1H, H-2). ES-MS: 281 [MH]$^+$.

EXAMPLE 151

2,4-Diamino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



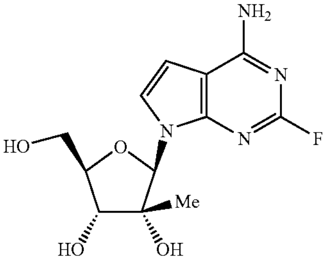
A mixture of the product from Step B of Example 118 (24 mg) in aqueous ammonia (30%, 10 mL) was heated in a stainless steel autoclave at 100° C. overnight, then cooled and evaporated. The residue was purified on a silica gel column with CH$_2$Cl$_2$/MeOH (10/1 and 5/1) as the eluent to afford the title compound (15 mg).

^{1}H NMR (DMSO-d$_6$): δ 0.68 (s, 3H, CH$_3$), 3.48–3.58 (m 1H, H-5'), 3.68–3.73 (m, 2H, H-5", H-4'), 3.84 (m, 1H, H-3'), 4.72 (s, 1H, 2'-OH), 4.97–5.03 (m, 2H, 3'-OH, 5'-OH), 5.45 (br s, 2H, NH$_2$), 6.00 (s, 1H, H-1'), 6.28 (d, 1H, J=3.7 Hz, H-5), 6.44 (br s, 2H, NH$_2$) 6.92 (d, 1H J=3.7 Hz, H-6).

ES MS: 294.1 (M–H$^+$).

EXAMPLE 152

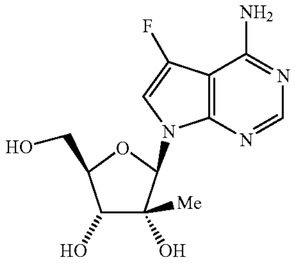
4-Amino-2-fluoro-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



To a solution of HF/pyridine (70%, 2 mL) diluted with pyridine (1 mL) at −30° C. is added the compound of Example 151 (60 mg, 0.2 mmol) in 0.5 mL pyridine followed by tert-butyl nitrite (36 μL, 0.3 mmol). Stirring is continued for 5 min −25° C. Then the solution is poured into ice-water (5 mL), neutralized with 2 N aqueous NaOH, and evaporated to dryness. The residue is purified on a silica gel column with CH$_2$Cl$_2$/MeOH (20/1 and 10/1) as the eluent to afford the title compound.

EXAMPLE 153

4-Amino-5-fluoro-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



Step A: 4-Acetylamino-7-(2,3,5-tri-O-acetyl-2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

To a solution of the compound from step F of Example 62 (280 mg, 1.00 mmol) in pyridine is added acetic anhydride (613 mg, 6.0 mmol). The resulting solution is stirred overnight at ambient temperature evaporated in vacuo and the resulting crude mixture is purified on silica gel using ethyl acetate/hexane as the eluent. Fractions containing the desired product are pooled and evaporated in vacuo to give the desired product.

Step B: 4-Acetylamino-5-bromo-7-(2,3,5-tri-O-acetyl-2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

To a pre-cooled (0° C.) solution of the compound from Step A (460 mg, 1.00 mmol) in DMF is added N-bromosuccinimide (178 mg, 1.0 mmol) in DMF. The resulting solution is stirred at 0° C. for 30 min then at room temperature for another 30 min. The reaction is quenched by addition of methanol and evaporated in vacuo. The resulting crude mixture is purified on silica gel using ethyl acetate/hexane as the eluent. Fractions containing the desired product are pooled and evaporated in vacuo to give the desired product.

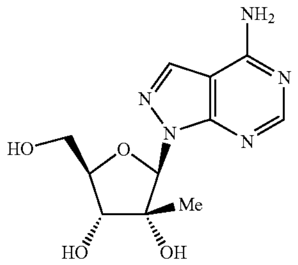
Step C: 4-Amino-5-fluoro-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

To a pre-cooled (−78° C.) solution of the compound from Step B (529 mg, 1.00 mmol) in THF is added butyl lithium

(2M in hexanes) (0.5 mL, 1.00 mmol). The resulting solution is stirred at −78° C. for 30 min and then quenched with N-fluorobenzensulfonimide (315 mg, 1.00 mmol) in THF. The resulting solution is very slowly allowed to come to ambient temperature and then poured into a stirred mixture of saturated aqueous ammonium chloride and dichloromethane. The organic phase is evaporated in vacuo and treated with ammonium hydroxide at 55° C. in a closed container overnight. The resulting crude mixture is purified on silica gel using dichloromethane/methanol as the eluent. Fractions containing the desired product are pooled and evaporated in vacuo to give the desired product.

EXAMPLE 154

4-Amino-1-(2-C-methyl-β-D-ribofuranosyl)-1H-pyrazolo[3,4-d]pyrimidine



Step A: 4-Amino-1-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C-methyl-β-D-ribofuranosyl]-1H-pyrazolo[3,4-d] pyrimidine

To the compound from Step C of Example 62 (1.00 g, 2.02 mmol) in dichloromethane (20 mL) was bubbled HBr gas for 5 min until it was saturated. The resulting solution was stirred at room temperature for 10 min, evaporated in vacuo and coevaporated with anhydrous toluene (10 mL). 4-Amino-1H-pyrazolo[3,4-d]pyrimidine (Aldrich, 0.43 g, 3.18 mmol) and NaH (60%, 150 mg, 3.8 mmol) were stirred in 1-methyl-2-pyrrolidinone (10 mL) for 30 min. The resulting solution was poured into the above bromo sugar residue and the mixture was stirred overnight. The mixture was diluted with toluene (50 mL), washed with brine (10%, 3×50 mL) and concentrated under reduced pressure. The residue was chromatographed on silica gel (EtOAc as eluent) to afford a solid (400 mg).

Step B: 4-Amino-1-(2-C-methyl-β-D-ribofuranosyl)-1H-pyrazolo[3,4-d]pyrimidine

To a solution of the compound from Step A (0.20 g, 0.33 mmol) in dichloromethane (10 mL) at −78° C. was added boron trichloride (1M in dichloromethane) (3 mL, 3 mmol) dropwise. The mixture was stirred at −78° C. for 0.5 h, then at −45° C. to −30° C. for 2 h. The reaction was quenched by addition of sodium acetate (1.0 g) and methanol (10 mL). The solution was evaporated and the residue was purified by flash chromatography over silica gel using CH_2Cl_2 and CH_2Cl_2/MeOH (95:5–90:10) gradient as the eluent to furnish the desired compound (60 mg) as a solid, which was recrystallized from methanol and acetonitrile to give the title compound as an off-white solid (40 mg).

^1H NMR (DMSO-d_6): δ 0.75 (s, 3H), 3.59 (m, 1H), 3.69 (m, 1H), 3.91 (m, 1H), 4.12 (m, 1H), 4.69 (t, 1H, J 5.1 Hz), 5.15 (m, 2H), 6.13 (s, 1H), 7.68 (s, br, 1H), 7.96 (s, br, 1H), 8.18 (s, 1H), 8.21 (s, 1H).

^{13}C NMR (DMSO-d_6): 19.32, 62.78, 74.11, 78.60, 83.65, 90.72, 99.79, 133.50, 153.89, 156.21, 158.05.

LC-MS: Found: 282.1 ($M+H^+$); calculated for $C_{11}H_{15}N_5O_4+H^+$. 282.1.

BIOLOGICAL ASSAYS

The assays employed to measure the inhibition of HCV NS5B polymerase and HCV replication are described below.

The effectiveness of the compounds of the present invention as inhibitors of HCV NS5B RNA-dependent RNA polymerase (RdRp) was measured in the following assay.

A. Assay for Inhibition of HCV NS5B Polymerase:

This assay was used to measure the ability of the nucleoside derivatives of the present invention to inhibit the enzymatic activity of the RNA-dependent RNA polymerase (NS5B) of the hepatitis C virus (HCV) on a heteromeric RNA template.

Procedure:

Assay Buffer Conditions: (50 μL -total/reaction)

20 mM Tris, pH 7.5
50 μM EDTA
5 mM DTT
2 mM $MgCl_2$
80 mM KCl
0.4 U/μL RNAsin (Promega, stock is 40 units/μL)
0.75 μg t500 (a 500-nt RNA made using T7 runoff transcription with a sequence from the NS2/3 region of the hepatitis C genome)
1.6 μg purified hepatitis C NS5B (form with 21 amino acids C-terminally truncated)
1 μM A,C,U,GTP (Nucleoside triphosphate mix)
[alpha-^{32}P]-GTP or [alpha-^{33}P]-GTP

The compounds were tested at various concentrations up to 100 μM final concentration.

An appropriate volume of reaction buffer was made including enzyme and template t500. Nucleoside derivatives of the present invention were pipetted into the wells of a 96-well plate. A mixture of nucleoside triphosphates (NTP's), including the radiolabeled GTP, was made and pipetted into the wells of a 96-well plate. The reaction was initiated by addition of the enzyme-template reaction solution and allowed to proceed at room temperature for 1–2 h.

The reaction was quenched by addition of 20 μL 0.5M EDTA, pH 8.0. Blank reactions in which the quench solution was added to the NTPs prior to the addition of the reaction buffer were included.

50 μL of the quenched reaction were spotted onto DE81 filter disks (Whatman) and allowed to dry for 30 min. The filters were washed with 0.3 M ammonium formate, pH 8 (150 mL/wash until the cpm in 1 mL wash is less than 100, usually 6 washes). The filters were counted in 5-mL scintillation fluid in a scintillation counter.

The percentage of inhibition was calculated according to the following

equation: % Inhibition=[1−(cpm in test reaction−cpm in blank)/(cpm in control reaction−cpm in blank)]×100.

Representative compounds tested in the HCV NS5B polymerase assay exhibited IC_{50}'s less than 100 micromolar.

B. Assay for Inhibition of HCV RNA Replication:

The compounds of the present invention were also evaluated for their ability to affect the replication of Hepatitis C Virus RNA in cultured hepatoma (HuH-7) cells containing a subgenomic HCV Replicon. The details of the assay are described below. This Replicon assay is a modification of that described in V. Lohmann, F. Korner, J-O. Koch, U. Herian, L. Theilmann, and R. Bartenschlager, "Replication of a Sub-genomic Hepatitis C Virus RNAs in a Hepatoma Cell Line," *Science* 285:110 (1999).

Protocol:

The assay was an in situ Ribonuclease protection, Scintillation Proximity based-plate assay (SPA). 10,000–40,000 cells were plated in 100–200 μL of media containing 0.8 mg/mL G418 in 96-well cytostar plates (Amersham). Compounds were added to cells at various concentrations up to 100 μM in 1% DMSO at time 0 to 18 h and then cultured for 24–96 h. Cells were fixed (20 min, 10% formalin), permeabilized (20 min, 0.25% Triton X-100/PBS) and hybridized (overnight, 50° C.) with a single-stranded ^{33}P RNA probe complementary to the (+) strand NS5B (or other genes) contained in the RNA viral genome. Cells were washed, treated with RNAse, washed, heated to 65° C. and counted in a Top-Count. Inhibition of replication was read as a decrease in counts per minute (cpm).

Human HuH-7 hepatoma cells, which were selected to contain a subgenomic replicon, carry a cytoplasmic RNA consisting of an HCV 5' non-translated region (NTR), a neomycin selectable marker, an EMCV IRES (internal ribosome entry site), and HCV non-structural proteins NS3 through NS5B, followed by the 3' NTR.

Representative compounds tested in the replication assay exhibited EC$_{50}$'s less than 100 micromolar.

The nucleoside derivatives of the present invention were also evaluated for cellular toxicity and anti-viral specificity in the counterscreens described below.

C. Counterscreens:

The ability of the nucleoside derivatives of the present invention to inhibit human DNA polymerases was measured in the following assays.

a. Inhibition of Human DNA Polymerases Alpha and Beta:

Reaction Conditions:

50 μL reaction volume

Reaction Buffer Components:

20 mM Tris-HCl, pH 7.5
200 μg/mL bovine serum albumin
100 mM KCl
2 mM β-mercaptoethanol
10 mM $MgCl_2$
1.6 μM dA, dG, dC, dTTP
α-^{33}P-dATP

Enzyme and Template:

0.05 mg/mL gapped fish sperm DNA template
0.01 U/μL DNA polymerase αor β

Preparation of Gapped Fish Sperm DNA Template:

Add 5 μL 1M $MgCl_2$ to 500 μL activated fish sperm DNA (USB 70076);
Warm to 37° C. and add 30 μL of 65 U/μL of exonuclease III (GibcoBRL 18013-011);
Incubate 5 min at 37° C.;
Terminate reaction by heating to 65° C. for 10 min;
Load 50–100 μL aliquots onto Bio-spin 6 chromatography columns (Bio-Rad 732-6002) equilibrated with 20 mM Tris-HCl, pH 7.5;
Elute by centrifugation at 1,000×g for 4 min;
Pool eluate and measure absorbance at 260 nm to determine concentration.

The DNA template was diluted into an appropriate volume of 20 mM Tris-HCl, pH 7.5 and the enzyme was diluted into an appropriate volume of 20 mM Tris-HCl, containing 2 mM 1B-mercaptoethanol, and 100 mM KCl. Template and enzyme were pipetted into microcentrifuge tubes or a 96 well plate. Blank reactions excluding enzyme and control reactions excluding test compound were also prepared using enzyme dilution buffer and test compound solvent, respectively. The reaction was initiated with reaction buffer with components as listed above. The reaction was incubated for 1 hour at 37° C. The reaction was quenched by the addition of 20 μL 0.5M EDTA. 50 μL of the quenched reaction was spotted onto Whatman DE81 filter disks and air dried. The filter disks were repeatedly washed with 150 mL 0.3M ammonium formate, pH 8 until 1 mL of wash is <100 cpm. The disks were washed twice with 150 mL absolute ethanol and once with 150 mL anhydrous ether, dried and counted in 5 mL scintillation fluid.

The percentage of inhibition was calculated according to the following

$$\text{equation: \% inhibition}=[1-(\text{cpm in test reaction}-\text{cpm in blank})/(\text{cpm in control reaction}-\text{cpm in blank})]\times 100.$$

b. Inhibition of Human DNA Polymerase Gamma:

The potential for inhibition of human DNA polymerase gamma was measured in reactions that included 0.5 μg/μL enzyme; 10 μM dATP, dGTP, dCTP, and TTP; 2 μCi/reaction [α-^{33}P]-dATP, and 0.4 μg/μL activated fish sperm DNA (purchased from US Biochemical) in a buffer containing 20 mM Tris pH8, 2 mM β-mercaptoethanol, 50 mM KCl, 10 mM MgCl2, and 0.1 μg/μL BSA. Reactions were allowed to proceed for 1 h at 37° C. and were quenched by addition of 0.5 M EDTA to a final concentration of 142 mM. Product formation was quantified by anion exchange filter binding and scintillation counting. Compounds were tested at up to 50 μM.

The percentage of inhibition was calculated according to the following

$$\text{equation: \% inhibition}=[1-(\text{cpm in test reaction}-\text{cpm in blank})/(\text{cpm in control reaction}-\text{cpm in blank})]\times 100.$$

The ability of the nucleoside derivatives of the present invention to inhibit HIV infectivity and HIV spread was measured in the following assays.

c. HIV Infectivity Assay

Assays were performed with a variant of HeLa Magi cells expressing both CXCR4 and CCR5 selected for low background β-galactosidase (β-gal) expression. Cells were infected for 48 h, and β-gal production from the integrated HIV-1 LTR promoter was quantified with a chemiluminescent substrate (Galactolight Plus, Tropix, Bedford, Mass.). Inhibitors were titrated (in duplicate) in twofold serial dilutions starting at 100 μM; percent inhibition at each concentration was calculated in relation to the control infection.

d. Inhibition of HIV Spread

The ability of the compounds of the present invention to inhibit the spread of the human immunedeficiency virus (HIV) was measured by the method described in U.S. Pat. No. 5,413,999 (May 9, 1995), and J. P. Vacca, et al., *Proc. Natl. Acad. Sci*., 91: 4096–4100 (1994), which are incorporated by reference herein in their entirety.

The nucleoside derivatives of the present invention were also screened for cytotoxicity against cultured hepatoma (HuH-7) cells containing a subgenomic HCV Replicon in an MTS cell-based assay as described in the assay below. The HuH-7 cell line is described in H. Nakabayashi, et al., *Cancer Res*., 42: 3858 (1982).

e. Cytotoxicity Assay:

Cell cultures were prepared in appropriate media at concentrations of approximately 1.5×10^5 cells/mL for suspension cultures in 3 day incubations and 5.0×10^4 cells/mL for adherent cultures in 3 day incubations. 99 μL of cell culture was transferred to wells of a 96-well tissue culture treated plate, and 1 μL of 100-times final concentration of the test compound in DMSO was added. The plates were incubated at 37° C. and 5% CO_2 for a specified period of time. After the incubation period, 20 μL of CellTiter 96 Aqueous One Solution Cell Proliferation Assay reagent (MTS) (Promega) was added to each well and the plates were incubated at 37° C. and 5% CO_2 for an additional period of time up to 3 h. The plates were agitated to mix well and absorbance at 490 nm was read using a plate reader. A standard curve of suspension culture cells was prepared with known cell numbers just prior to the addition of MTS reagent. Metabolically active cells reduce MTS to formazan. Formazan absorbs at 490 nm. The absorbance at 490 nm in the presence of compound was compared to absorbance in cells without any compound added. *Reference*: Cory, A. H. et al., "Use of an aqueous soluble tetrazolium/formazan assay for cell growth assays in culture," *Cancer Commun*. 3: 207 (1991).

The following assays were employed to measure the activity of the compounds of the present invention against other RNA-dependent RNA viruses:

a. Determination of In Vitro Antiviral Activity of Compounds Against Rhinovirus (Cytopathic Effect Inhibition Assay):

Assay conditions are described in the article by Sidwell and Huffman, "Use of disposable microtissue culture plates for antiviral and interferon induction studies," *Appl. Microbiol*. 22: 797–801 (1971).

Viruses:

Rhinovirus type 2 (RV-2), strain HGP, was used with KB cells and media (0.1% $NaHCO_3$, no antibiotics) as stated in the Sidwell and Huffman reference. The virus, obtained from the ATCC, was from a throat swab of an adult male with a mild acute febrile upper respiratory illness.

Rhinovirus type 9 (RV-9), strain 211, and rhinovirus type 14 (RV-14), strain Tow, were also obtained from the American Type Culture Collection (ATCC) in Rockville, Md. RV-9 was from human throat washings and RV-14 was from a throat swab of a young adult with upper respiratory illness. Both of these viruses were used with HeLa Ohio-1 cells (Dr. Fred Hayden, Univ. of VA) which were human cervical epitheloid carcinoma cells. MEM (Eagle's minimum essential medium) with 5% Fetal Bovine serum (FBS) and 0.1% $NaHCO_3$ was used as the growth medium.

Antiviral test medium for all three virus types was MEM with 5% FBS, 0.1% NaHCO3, 50 μg gentamicin/mL, and 10 mM $MgCl_2$.

2000 μg/mL was the highest concentration used to assay the compounds of the present invention. Virus was added to the assay plate approximately 5 min after the test compound. Proper controls were also run. Assay plates were incubated with humidified air and 5% Co_2 at 37° C. Cytotoxicity was monitored in the control cells microscopically for morphologic changes. Regression analysis of the virus CPE data and the toxicity control data gave the ED50 (50% effective dose) and CC50 (50% cytotoxic concentration). The selectivity index (SI) was calculated by the formula: SI=CC50÷ED50.

b. Determination of In Vitro Antiviral Activity of Compounds Against Dengue, Banzi, and Yellow Fever (CPE Inhibition Assay)

Assay details are provided in the Sidwell and Huffman reference above.

Viruses:

Dengue virus type 2, New Guinea strain, was obtained from the Center for Disease Control. Two lines of African green monkey kidney cells were used to culture the virus (Vero) and to perform antiviral testing (MA-104). Both Yellow fever virus, 17D strain, prepared from infected mouse brain, and Banzi virus, H336 strain, isolated from the serum of a febrile boy in South Africa, were obtained from ATCC. Vero cells were used with both of these viruses and for assay.

Cells and Media:

MA-104 cells (BioWhittaker, Inc., Walkersville, Md.) and Vero cells (ATCC) were used in Medium 199 with 5% FBS and 0.1% $NaHCO_3$ and without antibiotics.

Assay medium for dengue, yellow fever, and Banzi viruses was MEM, 2% FBS, 0.18% NaHCO3 and 50 μg gentamicin/mL.

Antiviral testing of the compounds of the present invention was performed according to the Sidwell and Huffman reference and similar to the above rhinovirus antiviral testing. Adequate cytopathic effect (CPE) readings were achieved after 5–6 days for each of these viruses.

c. Determination of In Vitro Antiviral Activity of Compounds Against West Nile Virus (CPE Inhibition Assay)

Assay details are provided in the Sidwell and Huffman reference cited above. West Nile virus, New York isolate derived from crow brain, was obtained from the Center for Disease Control. Vero cells were grown and used as described above. Test medium was MEM, 1% PBS, 0.1% NaHCO3 and 50 μg gentamicin/mL.

Antiviral testing of the compounds of the present invention was performed following the methods of Sidwell and Huffman which are similar to those used to assay for rhinovirus activity. Adequate cytopathic effect (CPE) readings were achieved after 5–6 days.

d. Determination of In Vitro Antiviral Activity of Compounds Against Rhino, Yellow Fever, Dengue, Banzi, and West Nile Viruses (Neutral Red Uptake Assay)

After performing the CPE inhibition assays above, an additional cytopathic detection method was used which is described in "Microtiter Assay for Interferon: Microspectrophotometric Quantitation of Cytopathic Effect," *Appl. Environ. Microbiol*. 31: 35–38 (1976). A Model EL309 microplate reader (Bio-Tek Instruments Inc.) was used to read the assay plate. ED50's and CD50's were calculated as above.

Example of a Pharmaceutical-Formulation

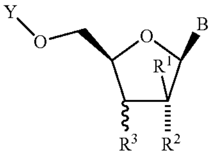
As a specific embodiment of an oral composition of a compound of the present invention, 50 mg of Example 61 or Example 62 is formulated with sufficient finely divided lactose to provide a total amount of 580 to 590 mg to fill a size O hard gelatin capsule.

While the invention has been described and illustrated in reference to specific embodiments thereof, those skilled in the art will appreciate that various changes, modifications, and substitutions can be made therein without departing from the spirit and scope of the invention. For example, effective dosages other than the preferred doses as set forth hereinabove may be applicable as a consequence of variations in the responsiveness of the human being treated for severity of the HCV infection. Likewise, the pharmacologic response observed may vary according to and depending upon the particular active compound selected or whether there are present pharmaceutical carriers, as well as the type of formulation and mode of administration employed, and such expected variations or differences in the results are contemplated in accordance with the objects and practices of the present invention. It is intended therefore that the invention be limited only by the scope of the claims which follow and that such claims be interpreted as broadly as is reasonable.

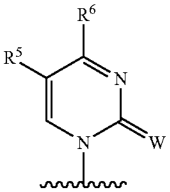
What is claimed is:

1. A method of treating hepatitis C virus (HCV) infection comprising administering to a mammal in need of such treatment a therapeutically effective amount of a compound of structural formula III, or a pharmaceutically acceptable salt or acyl derivatives thereof,

(III)



wherein B is



W is O or S;

Y is H, C_1-10 alkylcarbonyl, $P_3O_9H_4$, $P_2O_6H_3$, or P(O)R^9R^{10};

R^1 is CF_3, or C_{1-4} alkyl and one of R^2 and R^3 is OH or C_{1-4} alkoxy and the other of R^2 and R^3 is fluoro;

R^6 is H, OH, SH, NH_2, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, C_{3-6} cycloalkylamino, halogen, C_{1-4} alkyl, C_{1-4} alkoxy, or CF_3;

R^5 is H, C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-4} alkylamino, CF_3, or halogen; and

R^9 and R^{10} are each independently hydroxy, $OCH_2CH_2SC(=O)$t-butyl, or $OCH_2O(C=O)$iPr.

2. The method of claim **1** wherein a compound of structural formula III, or a pharmaceutically acceptable salt or acyl derivatives thereof is in combination with a therapeutic amount of another agent active against HCV infection selected from the group consisting of ribavirin; levovirin; thymosin apha-1; an inhibitor of NS3 serine protease; an inhibitor of inosine monophosphate dehydrogenase; and interferon-α or pegylated interferon-α.

* * * * *

UNITED STATES PATENT AND TRADEMARK OFFICE

CERTIFICATE OF CORRECTION

PATENT NO. : 7,105,499 B2
APPLICATION NO. : 10/250873
DATED : September 12, 2006
INVENTOR(S) : Steven S. Carroll et al.

Page 1 of 1

It is certified that error appears in the above-identified patent and that said Letters Patent is hereby corrected as shown below:

Claim 1, Column 138, Line 2, cancel "C_1-10 alkylcarbonyl" and substitute therefor -- C_{1-10} alkylcarbonyl --

Claim 2, Column 138, Line 23, cancel "apha-1" and substitute therefor -- alpha-1 --

Signed and Sealed this

Second Day of January, 2007

Jon W. Dudas

JON W. DUDAS
Director of the United States Patent and Trademark Office

US008481712B2

(12) **United States Patent**
Bhat et al.

(10) **Patent No.: US 8,481,712 B2**
(45) **Date of Patent: Jul. 9, 2013**

(54) **NUCLEOSIDE DERIVATIVES AS INHIBITORS OF RNA-DEPENDENT RNA VIRAL POLYMERASE**

(75) Inventors: **Balkrishen Bhat**, Carlsbad, CA (US); **Anne B. Eldrup**, Encinitas, CA (US); **Thazha P. Prakash**, Carlsbad, CA (US); **Phillip Dan Cook**, Fallbrook, CA (US); **Robert L. LaFemina**, Schwenksville, PA (US); **Amy L. Simcoe**, Collegeville, PA (US); **Carrie A. Rutkowski**, Lansdale, PA (US); **Mario A. Valenciano**, Scotch Plains, NJ (US)

(73) Assignees: **Merck Sharp & Dohme Corp.**, Rahway, NJ (US); **ISIS Pharmaceuticals, Inc.**, Carlsbad, CA (US)

(*) Notice: Subject to any disclaimer, the term of this patent is extended or adjusted under 35 U.S.C. 154(b) by 1830 days.

(21) Appl. No.: **11/701,682**

(22) Filed: **Feb. 2, 2007**

(65) **Prior Publication Data**

US 2012/0165515 A1 Jun. 28, 2012

Related U.S. Application Data

(60) Continuation of application No. 11/643,464, filed on Dec. 21, 2006, which is a continuation of application No. 11/200,499, filed on Aug. 9, 2005, now abandoned, which is a continuation of application No. 10/431,657, filed on May 7, 2003, now abandoned, which is a division of application No. 10/052,318, filed on Jan. 18, 2002, now Pat. No. 6,777,395.

(60) Provisional application No. 60/263,313, filed on Jan. 22, 2001, provisional application No. 60/282,069, filed on Apr. 6, 2001, provisional application No. 60/299,320, filed on Jun. 19, 2001, provisional application No. 60/344,528, filed on Oct. 25, 2001.

(51) **Int. Cl.**
C07H 19/10 (2006.01)
C07H 19/20 (2006.01)

(52) **U.S. Cl.**
USPC **536/26.2**; 536/26.26; 536/26.8

(58) **Field of Classification Search**
None
See application file for complete search history.

(56) **References Cited**

U.S. PATENT DOCUMENTS

3,480,613	A	11/1969	Walton	
1,187,824	A	4/1970	Walton	
4,211,773	A	7/1980	Lopez et al.	
4,666,892	A	5/1987	Fox et al.	
4,808,614	A	2/1989	Hertel	
5,672,594	A	9/1997	Weis et al.	
5,837,852	A *	11/1998	Chung et al.	536/24.5
5,977,061	A	11/1999	Holy et al.	
6,063,628	A	5/2000	Loeb et al.	
6,110,901	A	8/2000	Gluzman et al.	
6,348,587	B1	2/2002	Schinazi et al.	
6,777,395	B2 *	8/2004	Bhat et al.	514/43
6,784,161	B2	8/2004	Ismaili et al.	
6,812,219	B2	11/2004	LaColla et al.	
6,914,054	B2	7/2005	Sommadossi et al.	
7,101,861	B2	9/2006	Sommadossi et al.	
7,105,499	B2	9/2006	Carroll et al.	
7,112,600	B1	9/2006	Hashimoto et al.	
7,125,855	B2	10/2006	Bhat et al.	
7,157,441	B2	1/2007	Sommadossi et al.	
7,169,766	B2	1/2007	Sommadossi et al.	
7,202,224	B2	4/2007	Eldrup et al.	
7,429,572	B2	9/2008	Clark	
2002/0019363	A1	2/2002	Ismaili et al.	
2002/0053096	A1	5/2002	Hirschberg et al.	
2002/0055483	A1	5/2002	Watanabe et al.	
2002/0147160	A1	10/2002	Bhat et al.	
2002/0198171	A1	12/2002	Schinazi et al.	
2003/0007946	A1	1/2003	Narang et al.	
2003/0013089	A1	1/2003	Fisher et al.	
2003/0050229	A1	3/2003	Sommadossi et al.	
2003/0060400	A1	3/2003	LaColla et al.	
2004/0063622	A1	4/2004	Sommadossi et al.	
2004/0072788	A1	4/2004	Bhat et al.	
2004/0097461	A1	5/2004	Sommadossi et al.	
2004/0102414	A1	5/2004	Sommadossi et al.	

(Continued)

FOREIGN PATENT DOCUMENTS

FR	1.581.628	9/1969
GB	1209 654	10/1970

(Continued)

OTHER PUBLICATIONS

Mehta et al., Biochemistry (1976), 15(19), pp. 4329-4333.*
AN 1990:135032 for Savochkina et al., Molekulyarnaya Biologiya (Moscow), 1989, 23(6).*
Beers et al. "The Merck Manual of Diagnosis and Therapy", 1999, Seventeenth Edition, pp. 377-386, 1132, 1280.

(Continued)

Primary Examiner — Traviss C McIntosh, III
(74) *Attorney, Agent, or Firm* — Sheldon O. Heber; Jeffrey P. Bergman

(57) **ABSTRACT**

The present invention provides nucleoside compounds and certain derivatives thereof which are inhibitors of RNA-dependent RNA viral polymerase. These compounds are inhibitors of RNA-dependent RNA viral replication and are useful for the treatment of RNA-dependent RNA viral infection. They are particularly useful as inhibitors of hepatitis C virus (HCV) NS5B polymerase, as inhibitors of HCV replication, and/or for the treatment of hepatitis C infection. The invention also describes pharmaceutical compositions containing such nucleoside compounds alone or in combination with other agents active against RNA-dependent RNA viral infection, in particular HCV infection. Also disclosed are methods of inhibiting RNA-dependent RNA polymerase, inhibiting RNA-dependent RNA viral replication, and/or treating RNA-dependent RNA viral infection with the nucleoside compounds of the present invention.

11 Claims, No Drawings

U.S. PATENT DOCUMENTS

2005/0137161	A1	6/2005	Sommadossi et al.
2005/0272676	A1	12/2005	Bhat et al.
2006/0205686	A1	9/2006	Bhat et al.
2007/0275912	A1	11/2007	Bhat et al.
2010/0316594	A1	12/2010	Sommadossi et al.

FOREIGN PATENT DOCUMENTS

WO	WO 93/18051		9/1993
WO	WO 94/05687		3/1994
WO	98/16184	A2	4/1998
WO	WO 99/43691		9/1999
WO	WO 01/32153		5/2001
WO	WO 01/60315		8/2001
WO	WO 01/79246		10/2001
WO	WO 01/90121		11/2001
WO	WO 01/92282		12/2001
WO	WO 02/18404		3/2002
WO	0232920	A2	4/2002
WO	WO 02/48165	A2	6/2002
WO	WO 02/48165	A3	6/2002
WO	WO 02/069903	A2	9/2002
WO	WO 02/069903	A3	9/2002
WO	WO 02/094289	A1	11/2002
WO	WO 02/100354	A2	12/2002
WO	WO 02/100354	A3	12/2002
WO	WO 02/100415	A2	12/2002
WO	WO 03/000200	A2	1/2003
WO	WO 03/000713	A1	1/2003
WO	WO 03/015798	A1	2/2003
WO	WO 03/026589	A2	4/2003
WO	WO 03/026675	A1	4/2003
WO	WO 03/051881		6/2003
WO	WO 03/051896		6/2003
WO	WO 03/051897		6/2003
WO	WO 03/051898		6/2003
WO	WO 03/051899		6/2003
WO	WO 03/062255		7/2003
WO	03093290	A2	11/2003
WO	2004/003000	A2	1/2004

OTHER PUBLICATIONS

Chu et al. "9-Deazaadenosine—A New Potent Antitumor Agent", Biochemical Pharmacology, 1984, vol. 33, pp. 1229-1234.

Clark, et al. "Synthesis and antiviral activity of 2'-deoxy-2'-fluoro-2'-C-methyl purine nucleosides as inhibitors of hepatitis C virus RNA replication", Bioorganic & Medicinal Chemistry Letters, 2006, vol. 16, pp. 1712-1715.

Dee Nord et al. "Inhibition of Orotidylate Decarboxylase by 4(5H)-Oxo-1-B-D-Ribofuranosylpyrazolo[3,4-d] Pyrimidine-3-Thiocarboxamide (APR-TC) in B Lymphoblasts", Biochemical Pharmacology, 1988, vol. 37, pp. 4697-4705.

Eldrup et al. "Structure—Activity Relationship of Purine Ribonucleosides for Inhibition of Hepatitis C Virus RNA-Dependent RNA Polymerase", Journal of Medicinal Chemistry, 2004, vol. 47, pp. 2283-2295.

Flockhart, et al. "ATP analog specificity of cAMP-dependent protein kinase, cGMP-dependent protein kinase, and phosphorylase kinase", European Journal of Biochemistry, 1984, vol. 140, pp. 289-295.

Gupta et al. "Genetic and Biochemical Studies on Mutants of CHO Cells Resistant to 7-Deazapurine Nucleosides: Differences in the Mechanisms of Action of Toyocamycin and Tubercidin", Biochemical and Biophysical Research Communications, 1984, vol. 120, pp. 88-95.

Hoofnagle, "Therapy of Viral Hepatitis", Digestion, 1998, vol. 59, pp. 563-578.

Olsen, et al. A 7-Deaza-Adenosine Analog Is a Potent and Selective Inhibitor of Hepatitis C Virus Replication with Excellent Pharmacokinetic Properties, Antimicrobial Agents and Chemotherapy, 2004, vol. 48, pp. 3944-3953.

Ono et al. "Oligodeoxynucleotides containing 7-deazaadenine: synthesis and recognition by restriction endonucleases", Eleventh Symposium on Nucleic Acids Chemistry, Tokyo, Japan, Nov. 1-2, 1983, published in Nucleic Acids Symposium Series No. 12, pp. 67-70.

Seela, et al. "7-(B-D-Arabinofuranosyl)-2,4-dichlor-7H-pyrrolo[2,3-d]pyrimidin—Synthese, selektiver Halogenaustausch und EinfluB glyconischer Schutzgruppen auf die Reaktivitat des Aglycons", Liebigs Annalen der Chemie, 1984, pp. 722-733.

Smee et al. "Novel Pyrazolo[3,4-d]Pyrimidine Nucleoside Analog with Broad-Spectrum Antiviral Activity", Antimicrobial Agents and Chemotherapy, 1987, vol. 31, pp. 1535-1541.

Wolf et al. "New 2'-C-Branched-Chain Sugar Nucleoside Analogs with Potential Antiviral or Antitumor Activity", Synthesis, 1992, pp, 773-778.

HCAPLUS Accession No. 1958:89150, Abstract for Yukino, "Mouse hepatitis virus. II. Histopathological studies of infected mouse liver with correlation to the level of blood sugar and liver glycogen", Virus (Osaka), 1958, 8, pp. 67-72.

Bailly et al. "Treatment of HCV liver disease by recombinant interferon alpha", Nephrology Dialysis Transplantation, 1996, vol. 11, Suppl. 4, pp. 56-57.

Walton, E. et al., "Branched-Chain Sugar Nucleosides. V. Synthesis and Antiviral Properties of Serveral Branched-Chain Sugar Nucleosides" J. Med. Chem., vol. 12, pp. 306-309 (1969).

Matsuda, A. et al., "Radical Deoxygenation of Tert-Alcohols in 2'-Branched-Chain Sugar Pyrimidine Nucleosides: Synthesis and Antileukemic Acitivity of 2'-Deoxy-2'(S)-Methylcytidine", Chem. Pharm. Bull., vol. 35, pp. 3967-3970 (1987).

Matsuda, A. et al., Alkyl Addition Reaction of Pyrimidine 2'-Ketonucleosides: Synthesis of 2'-Branched-Chain Sugar Pyrimidine Nucleosides (Nucleosides and Nucleotides. LXXXI), Chem. Phar. Bull., vol. 36, pp. 945-953 (1988).

Murai, Y. et al., "Asynthesis and an X-Ray Analysis of 2'-C-, 3'-C-and 5'-C-Methylsangivamycins", Heterocycles, vol. 33, pp. 391-404 (1992).

Limori, T. et al., "2-C-, 3'-C-and 5'-C-Methylsangivamycins: Conformational Lock with the Methyl Group", Tetrahedron Letters, vol. 3, No. 49, pp. 7273-7276 (1991).

Wolfe, M. et al., "AConsis Synthesis of 2'-C-Methylribonucleosides", Tetrahedron Letters, vol. 36, No. 42, pp. 7611-7614 (1995).

Harry-O'kuru, R. et al., "Ashor, Flexible Route toward 2'-C-Branched Ribonucleosides", J. Org. Chem, vol. 62, pp. 1754-1759 (1997).

Gallo, M. et al., "Synthesis of 2'-Modified Nucleotides", Molecules, vol. 5, pp. 727-729 (2000).

Beigelman, L. et al., "new synthesis of 2'-C-Methylnucleosides Starting From D-Glucose and D-Ribose", Carbohydrate Research, vol. 166, pp. 219-232 (1987).

Anzai, K. et al., "A New Antibiotic, Tubercidin", The Journal of Antibiotics., vol. X, No. 5, pp. 201-205 (1957).

Tolman, R. et al., "Pyrrolopyrimidine Nucleosides. III. The Total Synthesis of Toyocamycin, Sangivamycin, Tubercindin, and Related Derivatives", Journal of the American Chemical Society, vol. 91, No. 8, pp. 2102-2108 (1969).

Tolman, R. et al., "Pyrrolo[2,3-d]pyrimidine Nucleoside Antiobiotics. Total Synthesis and Structure of toyacamycin, Unamycin B, Vengicide, Antibiotic E-212, and Sangivamycin (BA-90212)", Journal of the American Chemical Society, vol. 90, No. 2, pp. 24-526 (1968).

Siddiqi, S. et al., "Search for New Purine-and ribose-Modified Adenosine Analogues as Selective Agonists and Antagonists at Adenosine Receptors", J. Med. Chem., vol. 38, pp. 1174-1188 (1995).

Frachetti, P. et al., 2'-C-Methyl Analogues of Selective Adenosine receptor Agonists: Synthesis and Binding Studies:, J. Med. Chem, vol. 41, pp. 1708-1715 (1988).

Kalinichenko, E. et al., "Substrate Specificity of Adenosine Deminase. Role of Mehtyl Groups at the 2'-, 3', and 5' Carbon Atoms of Adenosine", Bioorganicheskaya Khimiya, vol. 14, No. 9, pp. 1157-1161 (1988).

Wolf, et al., "new 2'-c-Branched-Chain Suar Nucleoside Analogs with Potential Antiviral or Antitumor Activity", Synthesis, pp. 773-778 (Aug. 1992).

CAPLUS Database, Accession No. 129:156753, Abstract of Ilyin et al., "Ribavirin inhibits protein synthesis and cell proliferation induced by mitogenic factors in primary human and rat hepatocytes", Hepatology (Philadelphia) (1998), vol. 27, pp. 1687-1694.

CAPLUS Database, Accession No. 1980:599421, Abstract of Harada et al., "The nucleotide sequence of nuclear 4.8S RNA of mouse cells", Biochem. Biophys. Res. Commun. (1980), vol. 95, pp. 1332-1340.

Abbruzzese, James L. et al., Phase I trial of 1-(2'-deoxy-2' -fluoro-1-beta-D-arabinofuranosyl)-5-methyluracil (FMAU) terminated by severe neurologic toxicity, Investigational New Drugs, 1989, vol. 7, pp. 195-201.

Chu, Chung, K. et al., "Use of 2'-Fluoro-5-Methyl-β-L-Arabinofuranosyluracil as a Novel Antiviral Agent for Hepatitis B Virus and Epstein-Barr Virus", Antimicrobial Agents and Chemotherapy, 1995, vol. 39, No. 4., pp. 979-981.

Harada, Kazuho et al., "Nucleosides. 139. Synthesis and Anticytomegalovirus and Antiherpes Simplex Virus Activity of 5'-Modified Analogues of 2'-Fluoroarabinosylpyrimidine Nucleosides", J. Med. Chem., 1987, vol. 30, pp. 226-229.

Merluzzi, V.J. et al., "Comparison of 2'-Fluoro-Arabinosyl Pyrimidine Nucleosides and 1-β-D-Arabinofuranosylcytosine on Immunological Parameters in Vitro", Int. J. Immunopharmacology, 1983, vol. 5, No. 5, pp. 421-425.

Zoulim, Fabien et al., 2',3'-Dideoxy-β-L-5-Fluorocytidine Inhibits Duck Hepatitis B Virus Reverse Transcription and Suppresses Viral DNA Synthesis in Hepatocytes, both in Vitro and in Vivo, Antimicrobial Agents and Chemotherapy, 1996, vol. 40, No. 2., pp. 448-453.

* cited by examiner

NUCLEOSIDE DERIVATIVES AS INHIBITORS OF RNA-DEPENDENT RNA VIRAL POLYMERASE

CROSS-REFERENCE TO RELATED APPLICATIONS

This application is a continuation of application Ser. No. 11/643,464, filed Dec. 21, 2006, copending herewith, which is a continuation of application Ser. No. 11/200,499, filed Aug. 9, 2005, now abandoned, which is a continuation of application Ser. No. 10/431,657, filed May 7, 2003, now abandoned, which is a Division of application Ser. No. 10/052,318, filed Jan. 18, 2002, issued as U.S. Pat. No. 6,777,395 on Aug. 17, 2004, which claims priority under 35 U.S.C. §119(e) from provisional application Ser. Nos. 60/263,313, filed Jan. 22, 2001; 60/282,069, filed Apr. 6, 2001; 60/299,320, filed Jun. 19, 2001; and 60/344,528, filed Oct. 25, 2001; the contents of each of which are hereby incorporated by reference.

FIELD OF THE INVENTION

The present invention provides nucleoside compounds and certain derivatives thereof which are inhibitors of RNA-dependent RNA viral polymerase. These compounds are inhibitors of RNA-dependent RNA viral replication and are useful for the treatment of RNA-dependent RNA viral infection. They are particularly useful as inhibitors of hepatitis C virus (HCV) NS5B polymerase, as inhibitors of HCV replication, and for the treatment of hepatitis C infection.

BACKGROUND OF THE INVENTION

Hepatitis C virus (HCV) infection is a major health problem that leads to chronic liver disease, such as cirrhosis and hepatocellular carcinoma, in a substantial number of infected individuals, estimated to be 2-15% of the world's population. There are an estimated 4.5 million infected people in the United States alone, according to the U.S. Center for Disease Control. According to the World Health Organization, there are more than 200 million infected individuals worldwide, with at least 3 to 4 million people being infected each year. Once infected, about 20% of people clear the virus, but the rest harbor HCV the rest of their lives. Ten to twenty percent of chronically infected individuals eventually develop liver-destroying cirrhosis or cancer. The viral disease is transmitted parenterally by contaminated blood and blood products, contaminated needles, or sexually and vertically from infected mothers or carrier mothers to their off-spring. Current treatments for HCV infection, which are restricted to immunotherapy with recombinant interferon-α alone or in combination with the nucleoside analog ribavirin, are of limited clinical benefit. Moreover, there is no established vaccine for HCV. Consequently, there is an urgent need for improved therapeutic agents that effectively combat chronic HCV infection. The state of the art in the treatment of HCV infection has been reviewed, and reference is made to the following publications: B. Dymock, et al., "Novel approaches to the treatment of hepatitis C virus infection," *Antiviral Chemistry & Chemotherapy,* 11: 79-96 (2000); H. Rosen, et al., "Hepatitis C virus: current understanding and prospects

for future therapies," *Molecular Medicine Today,* 5: 393-399 (1999); D. Moradpour, et al., "Current and evolving therapies for hepatitis C," *European J. Gastroenterol. Hepatol.,* 11: 1189-1202 (1999); R. Bartenschlager, "Candidate Targets for Hepatitis C Virus-Specific Antiviral Therapy," *Intervirology,* 40: 378-393 (1997); G. M. Lauer and B. D. Walker, "Hepatitis C Virus Infection," *N. Engl. J. Med.,* 345: 41-52 (2001); B. W. Dymock, "Emerging therapies for hepatitis C virus infection," *Emerging Drugs,* 6: 13-42 (2001); and C. Crabb, "Hard-Won Advances Spark Excitement about Hepatitis C," *Science:* 506-507 (2001); the contents of all of which are incorporated by reference herein in their entirety.

Different approaches to HCV therapy have been taken, which include the inhibition of viral serine proteinase (NS3 protease), helicase, and RNA-dependent RNA polymerase (NS5B), and the development of a vaccine.

The HCV virion is an enveloped positive-strand RNA virus with a single oligoribonucleotide genomic sequence of about 9600 bases which encodes a polyprotein of about 3,010 amino acids. The protein products of the HCV gene consist of the structural proteins C, E1, and E2, and the non-structural proteins NS2, NS3, NS4A and NS4B, and NS5A and NS5B. The nonstructural (NS) proteins are believed to provide the catalytic machinery for viral replication. The NS3 protease releases NS5B, the RNA-dependent RNA polymerase from the polyprotein chain. HCV NS5B polymerase is required for the synthesis of a double-stranded RNA from a single-stranded viral RNA that serves as a template in the replication cycle of HCV. NS5B polymerase is therefore considered to be an essential component in the HCV replication complex [see K. Ishi, et al., "Expression of Hepatitis C Virus NS5B Protein: Characterization of Its RNA Polymerase Activity and RNA Binding," *Hepatology,* 29: 1227-1235 (1999) and V. Lohmann, et al., "Biochemical and Kinetic Analyses of NS5B RNA-Dependent RNA Polymerase of the Hepatitis C Virus," *Virology,* 249: 108-118 (1998)]. Inhibition of HCV NS5B polymerase prevents formation of the double-stranded HCV RNA and therefore constitutes an attractive approach to the development of HCV-specific antiviral therapies.

It has now been found that nucleoside compounds of the present invention and certain derivatives thereof are potent inhibitors of RNA-dependent RNA viral replication and in particular HCV replication. The 5'-triphosphate derivatives of the nucleoside compounds are inhibitors of RNA-dependent RNA viral polymerase and in particular HCV NS5B polymerase. The instant nucleoside compounds and derivatives thereof are useful to treat RNA-dependent RNA viral infection and in particular HCV infection.

It is therefore an object of the present invention to provide nucleoside compounds and certain derivatives thereof which are useful as inhibitors of RNA-dependent RNA viral polymerase and in particular as inhibitors of HCV NS5B polymerase.

It is another object of the present invention to provide nucleoside derivatives which are useful as inhibitors of the replication of an RNA-dependent RNA virus and in particular as inhibitors of the replication of hepatitis C virus.

It is another object of the present invention to provide nucleoside compounds and certain derivatives which are useful in the treatment of RNA-dependent RNA viral infection and in particular in the treatment of HCV infection.

It is another object of the present invention to provide pharmaceutical compositions comprising the novel compounds of the present invention in association with a pharmaceutically acceptable carrier.

It is another object of the present invention to provide pharmaceutical compositions comprising the nucleoside compounds and derivatives thereof for use as inhibitors of RNA-dependent RNA viral polymerase and in particular as inhibitors of HCV NS5B polymerase.

It is another object of the present invention to provide pharmaceutical compositions comprising the nucleoside compounds and derivatives thereof for use as inhibitors of RNA-dependent RNA viral replication and in particular as inhibitors of HCV replication.

It is another object of the present invention to provide pharmaceutical compositions comprising the nucleoside compounds and derivatives thereof for use in the treatment of RNA-dependent RNA viral infection and in particular in the treatment of HCV infection.

It is another object of the present invention to provide pharmaceutical compositions comprising the nucleoside compounds and derivatives thereof in combination with other agents active against an RNA-dependent RNA virus and in particular against HCV.

It is another object of the present invention to provide methods for the inhibition of RNA-dependent RNA viral polymerase and in particular for the inhibition of HCV NS5B polymerase.

It is another object of the present invention to provide methods for the inhibition of RNA-dependent RNA viral replication and in particular for the inhibition of HCV replication.

It is another object of the present invention to provide methods for the treatment of RNA-dependent RNA viral infection and in particular for the treatment of HCV infection.

It is another object of the present invention to provide methods for the treatment of RNA-dependent RNA viral infection in combination with other agents active against RNA-dependent RNA virus and in particular for the treatment of HCV infection in combination with other agents active against HCV.

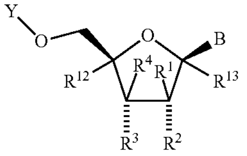
It is another object of the present invention to provide nucleoside compounds and certain derivatives thereof and their pharmaceutical compositions for use as a medicament for the inhibition of RNA-dependent RNA viral replication and/or the treatment of RNA-dependent RNA viral infection and in particular for the inhibition of HCV replication and/or the treatment of HCV infection.

It is another object of the present invention to provide for the use of the nucleoside compounds and certain derivatives thereof of the present invention and their pharmaceutical compositions for the manufacture of a medicament for the inhibition of RNA-dependent RNA viral replication and/or the treatment of RNA-dependent RNA viral infection and in particular for the inhibition of HCV replication and/or the treatment of HCV infection.

These and other objects will become readily apparent from the detailed description which follows.

SUMMARY OF THE INVENTION

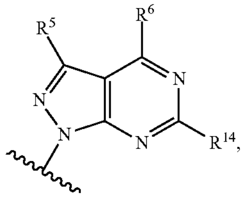
The present invention provides a method for inhibiting RNA-dependent RNA viral polymerase, a method for inhibiting RNA-dependent RNA viral replication, and/or a method for treating RNA-dependent viral infection in a mammal in need thereof, comprising administering to the mammal a therapeutically effective amount of a compound of structural formula I which is of the stereochemical configuration:

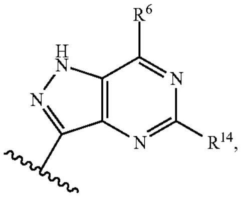


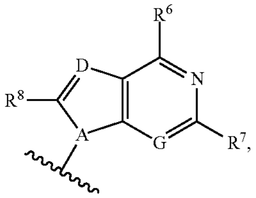
(I)

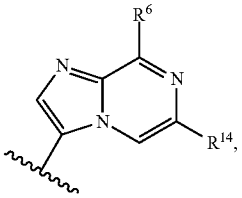
or a pharmaceutically acceptable salt thereof;

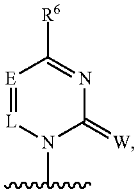
wherein B is selected from the group consisting of

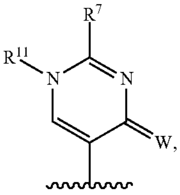












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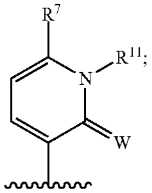


A, G, and L are each independently CH or N;

D is N, CH, C—CN, C—NO_2, C—C_{1-3} alkyl, C—$NHCONH_2$, C—$CONR^{11}R^{11}$, C—$CSNR^{11}R^{11}$, C—$COOR^{11}$, C—C(=NH)NH_2, C-hydroxy, C—C_{1-3} alkoxy, C-amino, C—C_{1-4} alkylamino, C-di(C_{1-4} alkyl)amino, C-halogen, C-(1,3-oxazol-2-yl), C-(1,3-thiazol-2-yl), or C-(imidazol-2-yl); wherein alkyl is unsubstituted or substituted with one to three groups independently selected from halogen, amino, hydroxy, carboxy, and C_{1-3} alkoxy;

E is N or CR^5;

W is O or S;

Y is H, C_{1-10} alkylcarbonyl, $P_3O_9H_4$, $P_2O_6H_3$, or $P(O)R^9R^{10}$;

R^1 is hydrogen, C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms and one of R^2 and R^3 is hydroxy or C_{1-4} alkoxy and the other of R^2 and R^3 is selected from the group consisting of
- hydrogen,
- hydroxy,
- halogen,
- C_{1-4} alkyl, optionally substituted with 1 to 3 fluorine atoms,
- C_{1-10} alkoxy, optionally substituted with C_{1-3} alkoxy or 1 to 3 fluorine atoms,

C_{2-6} alkenyloxy,
C_{1-4} alkylthio,
C_{1-8} alkylcarbonyloxy,
aryloxycarbonyl,
azido,
amino,
C_{1-4} alkylamino, and
di(C_{1-4} alkyl)amino; or

R^2 is hydrogen, C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms and one of R^1 and R^3 is hydroxy or C_{1-4} alkoxy and the other of R^1 and R^3 is selected from the group consisting of
hydrogen,
hydroxy,
halogen,
C_{1-4} alkyl, optionally substituted with 1 to 3 fluorine atoms,
C_{1-10} alkoxy, optionally substituted with hydroxy, C_{1-3} alkoxy, carboxy, or 1 to 3 fluorine atoms,
C_{2-6} alkenyloxy,
C_{1-4} alkylthio,
C_{1-8} alkylcarbonyloxy,
aryloxycarbonyl,
azido,
amino,
C_{1-4} alkylamino, and
di(C_{1-4} alkyl)amino; or

R^1 and R^2 together with the carbon atom to which they are attached form a 3- to 6-membered saturated monocyclic ring system optionally containing a heteroatom selected from O, S, and NC_{0-4} alkyl;

R^4 and R^6 are each independently H, OH, SH, NH_2, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, C_3-6 cycloalkylamino, halogen, C_{1-4} alkyl, C_{1-4} alkoxy, or CF_3;

R^5 is H, C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-4} alkylamino, CF_3, or halogen;

R^{14} is H, CF_3, C_{1-4} alkyl, amino, C_{1-4} alkylamino, C_{3-6} cycloalkylamino, or di(C_{1-4} alkyl)amino;

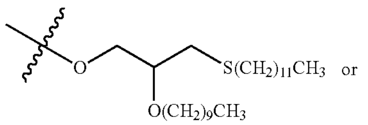
R^7 is hydrogen, amino, C_{1-4} alkylamino, C_{3-6} cycloalkylamino, or di(C_{1-4} alkyl)amino;

each R^{11} is independently H or C_{1-6} alkyl;

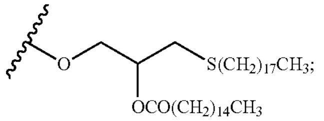
R^8 is H, halogen, CN, carboxy, C_{1-4} alkyloxycarbonyl, N_3, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, hydroxy, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} alkylsulfonyl, or (C_{1-4} alkyl)$_{0-2}$ aminomethyl;

R^{12} and R^{13} are each independently hydrogen, methyl, hydroxymethyl, or fluoromethyl; and

R^9 and R^{10} are each independently hydroxy, $OCH_2CH_2SC({=}O)C_{1-4}$ alkyl, $OCH_2O(C{=}O)OC_{1-4}$ alkyl, NHCHMeCO$_2$Me, OCH(C_{1-4} alkyl)O(C=O)C_{1-4} alkyl,

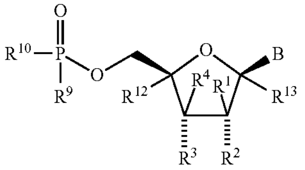


or



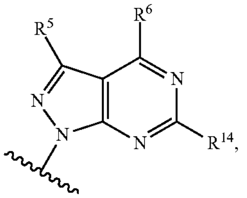
with the provisos that (a) when R^1 is hydrogen, one of R^3 and R^4 is hydrogen, and R^2 is fluoro, then the other of R^3 and R^4 is not hydrogen, halogen, azido, trifluoromethyl, C_{1-4} alkyl, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, or C_{1-10} alkoxy; (b) when R^1 is hydrogen, one of R^3 and R^4 is hydrogen, and R^2 is halogen, hydroxy, C_{1-6} alkoxy, or C_{2-6} alkenyloxy, then the other of R^3 and R^4 is not hydrogen, fluoro, or azido; and (c) when R^1 and R^3 are hydrogen and R^2 is hydroxy, then R^4 is not hydroxy.

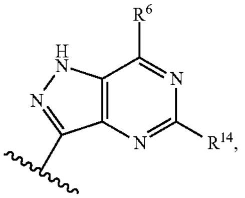
The present invention also provides novel compounds of structural formula IV of the indicated stereochemical configuration which are useful as inhibitors of RNA-dependent RNA viral polymerase. The compounds of formula IV are also inhibitors of RNA-dependent RNA viral replication and are useful for the treatment of RNA-dependent RNA viral infection:

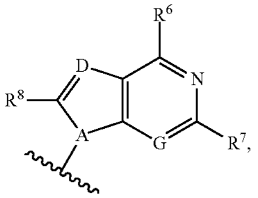


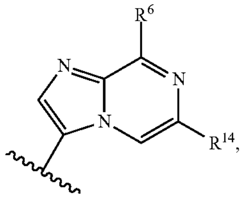
(IV)

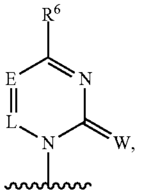
wherein B is selected from the group consisting of

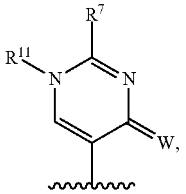












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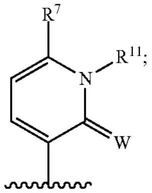


A, G, and L are each independently CH or N;

D is N, CH, C—CN, C—NO_2, C—C_{1-3} alkyl, C—NHCONH$_2$, C—CONR^{11}R^{11}, C—CSNR^{11}R^{11}, C—COOR^{11}, C—C(=NH)NH$_2$, C-hydroxy, C—C_{1-3} alkoxy, C-amino, C—C_{1-4} alkylamino, C-di(C_{1-4} alkyl)amino, C-halogen, C-(1,3-oxazol-2-yl), C-(1,3-thiazol-2-yl), or C-(imidazol-2-yl); wherein alkyl is unsubstituted or substituted with one to three groups independently selected from halogen, amino, hydroxy, carboxy, and C_{1-3} alkoxy;

E is N or CR^5;

W is O or S;

R^1 is hydrogen, C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms and one of R^2 and R^3 is hydroxy or C_{1-4} alkoxy and the other of R^2 and R^3 is selected from the group consisting of

hydrogen,
hydroxy,
halogen,
C_{1-4} alkyl, optionally substituted with 1 to 3 fluorine atoms,
C_{1-10} alkoxy, optionally substituted with C_{1-3} alkoxy or 1 to 3 fluorine atoms,
C_{2-6} alkenyloxy,
C_{1-4} alkylthio,
C_{1-8} alkylcarbonyloxy,
aryloxycarbonyl,
azido,
amino,
C_{1-4} alkylamino, and
di(C_{1-4} alkyl)amino; or

R^2 is hydrogen, C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms and one of R^1 and R^3 is hydroxy or C_{1-4} alkoxy and the other of R^1 and R^3 is selected from the group consisting of

hydrogen,
hydroxy,
halogen,
C_{1-4} alkyl, optionally substituted with 1 to 3 fluorine atoms,
C_{1-10} alkoxy, optionally substituted with hydroxy, C_{1-3} alkoxy, carboxy, or 1 to 3 fluorine atoms,
C_{2-6} alkenyloxy,
C_{1-4} alkylthio,
C_{1-8} alkylcarbonyloxy,
aryloxycarbonyl,
azido,
amino,
C_{1-4} alkylamino, and
di(C_{1-4} alkyl)amino; or

R^1 and R^2 together with the carbon atom to which they are attached form a 3- to 6-membered saturated monocyclic ring system optionally containing a heteroatom selected from O, S, and NC_{0-4} alkyl;

each R^4 is independently H, OH, SH, NH_2, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, C_{3-6} cycloalkylamino, halogen, C_{1-4} alkyl, C_{1-4} alkoxy, or CF_3;

R^4 and R^6 are each independently H, OH, SH, C_{1-4} alkylamino, alkyl)amino, C_{3-6} cycloalkylamino, halogen, C_{1-4} alkyl, C_{1-4} alkoxy, or CF_3;

R^5 is H, C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-4} alkylamino, CF_3, or halogen;

R^{14} is H, CF_3, C_{1-4} alkyl, amino, C_{1-4} alkylamino, C_{3-6} cycloalkylamino, or di(C_{1-4} alkyl)amino;

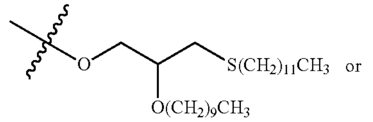
R^7 is hydrogen, amino, C_{1-4} alkylamino, C_{3-6} cycloalkylamino, or di(C_{1-4} alkyl)amino;

each R^{11} is independently H or C_{1-6} alkyl;

R^8 is H, halogen, CN, carboxy, C_{1-4} alkyloxycarbonyl, N_3, amino, C_{1-4} alkylamino, di(C_{1-4} alkylamino, hydroxy, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} alkylsulfonyl, or (C_{1-4} alkyl)$_{0-2}$ aminomethyl;

R^{12} and R^{13} are each independently hydrogen, methyl, hydroxymethyl, or fluoromethyl; and

R^9 and R^{10} are each independently hydroxy, $OCH_2CH_2SC({=}O)C_{1-4}$ alkyl, $OCH_2O(C{=}O)OC_{1-4}$ alkyl, $NHCHMeCO_2Me$, $OCH(C_{1-4}$ alkyl$)O(C{=}O)C_{1-4}$ alkyl,



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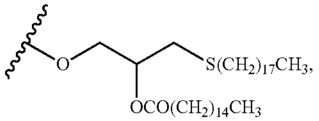
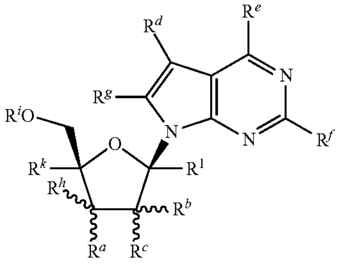


provided that at least one of R^9 and R^{10} is not hydroxy.

The present invention further provides novel compounds of structural formula XII of the indicated stereochemical configuration which are useful as inhibitors of RNA-dependent RNA viral polymerase and in particular of HCV NS5B polymerase:

(XII)



wherein R^a and R^h are each independently selected from the group consisting of hydrogen, cyano, azido, halogen, hydroxy, mercapto, amino, C_{1-4} alkoxy, C_{2-4} alkenyl, C_{2-4} alkynyl, and C_{1-4} alkyl, wherein alkyl is unsubstituted or substituted with hydroxy, amino, C_{1-4} alkoxy, C_{1-4} alkylthio, or one to three fluorine atoms;

R^b is C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl, wherein alkyl is unsubstituted or substituted with hydroxy, amino, C_{1-4} alkoxy, C_{1-4} alkylthio, or one to three fluorine atoms;

R^c is hydrogen, fluorine, hydroxy, mercapto, C_{1-4} alkoxy, or C_{1-4} alkyl; or R^b and R^c together with the carbon atom to which they are attached form a 3- to 6-membered saturated monocyclic ring system optionally containing a heteroatom selected from O, S, and NC_{0-4} alkyl;

R^d is hydrogen, cyano, nitro, C_{1-3} alkyl, $NHCONH_2$, CONRjRj, CSNRjRj, COORj, $C({=}NH)NH_2$, hydroxy, C_{1-3} alkoxy, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, halogen, (1,3-oxazol-2-yl), (1,3-thiazol-2-yl), or (imidazol-2-yl); wherein alkyl is unsubstituted or substituted with one to three groups independently selected from halogen, amino, hydroxy, carboxy, and C_{1-3} alkoxy;

R^e and R^f are each independently hydrogen, hydroxy, halogen, C_{1-4} alkoxy, amino, C_{1-4} alkylamino, di(C_{1-4} alkylamino, C_{3-6} cycloalkylamino, di(C_{3-6} cycloalkyl)amino, or C_{4-6} cycloheteroalkyl, unsubstituted or substituted with one to two groups independently selected from halogen, hydroxy, amino, C_{1-4} alkyl, and C_{1-4} alkoxy;

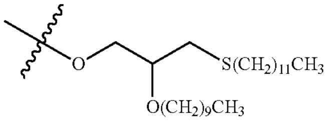
R^g is hydrogen, C_{1-4} alkyl, C_{2-4} alkynyl, halogen, cyano, carboxy, C_{1-4} alkyloxycarbonyl, azido, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, hydroxy, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} alkylsulfonyl, (C_{1-4} alkyl)$_{0-2}$ aminomethyl, or C_{4-6} cycloheteroalkyl, unsubstituted or substituted with one to two groups independently selected from halogen, hydroxy, amino, C_{1-4} alkyl, and C_{1-4} alkoxy;

R^i is hydrogen, C_{1-10} alkylcarbonyl, $P_3O_9H_4$, $P_2O_6H_3$, or $P(O)R^{m}R^{n}$;

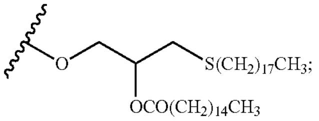
each R^j is independently hydrogen or C_{1-6} alkyl;

R^k and R^l are each independently hydrogen, methyl, hydroxymethyl, or fluoromethyl; and

R^m and R^n are each independently hydroxy, $OCH_2CH_2SC(=O)C_{1-4}$ alkyl, $OCH_2O(C=O)OC_{1-4}$ alkyl, $NHCHMeCO_2Me$, $OCH(C_{1-4}$ alkyl$)O(C=O)C_{1-4}$ alkyl,



or



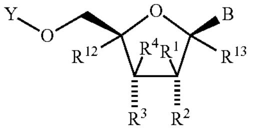
with the proviso that when R^a and R^c are α-hydroxy, R^e is amino, R^b is β-methyl and R^h is hydrogen or R^h is β-methyl and R^b is hydrogen, and R^f, R^g, R^i, R^k, and R^l are hydrogen, then R^d is not cyano or $CONH_2$.

The compounds of formula XII are also inhibitors of RNA-dependent RNA viral replication and in particular of HCV replication and are useful for the treatment of RNA-dependent RNA viral infection and in particular for the treatment of HCV infection.

Also encompassed within the present invention are pharmaceutical compositions containing the compounds alone or in combination with other agents active against RNA-dependent RNA virus and in particular against HCV.

DETAILED DESCRIPTION OF THE INVENTION

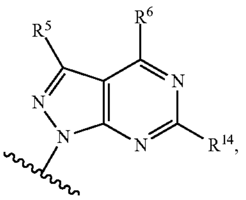
The present invention provides a method for inhibiting RNA-dependent RNA viral polymerase, a method for inhibiting RNA-dependent RNA viral replication, and/or a method for treating RNA-dependent RNA viral infection in a mammal in need thereof comprising administering to the mammal a therapeutically effective amount of a compound of structural formula I which is of the stereochemical configuration:

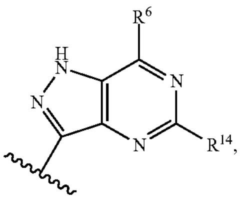


(I)

or a pharmaceutically acceptable salt thereof;

wherein B is selected from the group consisting of





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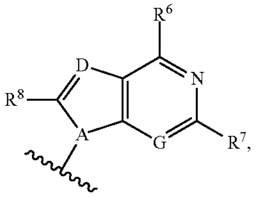
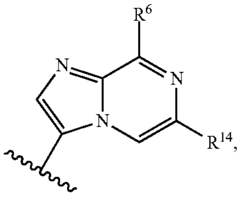
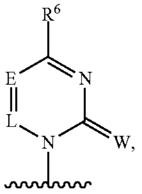
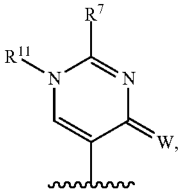
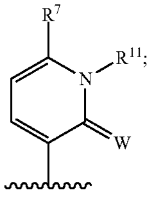








and



A, G, and L are each independently CH or N;

D is N, CH, C—CN, C—NO_2, C—C_{1-3} alkyl, C—$NHCONH_2$, C—$CONR^{11}R^{11}$, C—$CSNR^{11}R^{11}$, C—$COOR^{11}$, C—$C(=NH)NH_2$, C-hydroxy, C—C_{1-3} alkoxy, C-amino, C—C_{1-4} alkylamino, C-di(C_{1-4} alkyl)amino, C-halogen, C-(1,3-oxazol-2-yl), C-(1,3-thiazol-2-yl), or C-(imidazol-2-yl); wherein alkyl is unsubstituted or substituted with one to three groups independently selected from halogen, amino, hydroxy, carboxy, and C_{1-3} alkoxy;

E is N or CR^5;

W is O or S;

Y is H, C_{1-10} alkylcarbonyl, $P_3O_9H_4$, $P_2O_6H_3$, or $P(O)R^9R^{10}$;

R^1 is hydrogen, C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms and one of R^2 and R^3 is hydroxy or C_{1-4} alkoxy and the other of R^2 and R^3 is selected from the group consisting of
- hydrogen,
- hydroxy,
- halogen,
- C_{1-4} alkyl, optionally substituted with 1 to 3 fluorine atoms,
- C_{1-10} alkoxy, optionally substituted with C_{1-3} alkoxy or 1 to 3 fluorine atoms,
- C_{2-6} alkenyloxy,
- C_{1-4} alkylthio,
- C_{1-8} alkylcarbonyloxy,
- aryloxycarbonyl,
- azido,
- amino,
- C_{1-4} alkylamino, and
- di(C_{1-4} alkyl)amino; or

R^2 is hydrogen, C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms and one of R^1 and R^3 is hydroxy or C_{1-4} alkoxy and the other of R^1 and R^3 is selected from the group consisting of
- hydrogen,
- hydroxy,
- halogen,
- C_{1-4} alkyl, optionally substituted with 1 to 3 fluorine atoms,
- C_{1-10} alkoxy, optionally substituted with hydroxy, C_{1-3} alkoxy, carboxy, or 1 to 3 fluorine atoms,
- C_{2-6} alkenyloxy,
- C_{1-4} alkylthio,

C_{1-8} alkylcarbonyloxy,
aryloxycarbonyl,
azido,
amino,
C_{1-4} alkylamino, and
di(C_{1-4} alkyl)amino; or

R^1 and R^2 together with the carbon atom to which they are attached form a 3- to 6-membered saturated monocyclic ring system optionally containing a heteroatom selected from O, S, and NC_{0-4} alkyl;

R^4 and R^6 are each independently H, OH, SH, NH_2, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, C_{3-6} cycloalkylamino, halogen, C_{1-4} alkyl, C_{1-4} alkoxy, or CF_3;

R^5 is H, C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-4} alkylamino, CF_3, or halogen;

R^{14} is H, CF_3, C_{1-4} alkyl, amino, C_{1-4} alkylamino, C_{3-6} cycloalkylamino, or di(C_{1-4} alkyl)amino;

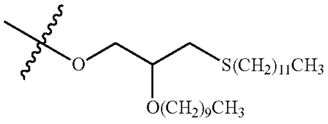
R^7 is hydrogen, amino, C_{1-4} alkylamino, C_{3-6} cycloalkylamino, or di(C_{1-4} alkyl)amino;

each R^{11} is independently H or C_{1-6} alkyl;

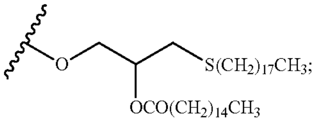
R^8 is H, halogen, CN, carboxy, C_{1-4} alkyloxycarbonyl, N_3, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, hydroxy, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} alkylsulfonyl, or (C_{1-4} alkyl)$_{0-2}$ aminomethyl;

R^{12} and R^{13} are each independently hydrogen, methyl, hydroxymethyl, or fluoromethyl; and

R^9 and R^{10} are each independently hydroxy, $OCH_2CH_2SC({=}O)C_{1-4}$ alkyl, $OCH_2O(C{=}O)OC_{1-4}$ alkyl, $NHCHMeCO_2Me$, $OCH(C_{1-4}$ alkyl$)O(C{=}O)C_{1-4}$ alkyl,

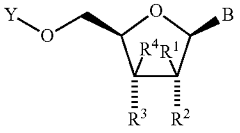


or



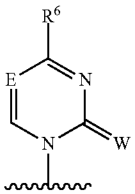
with the provisos that (a) when R^1 is hydrogen, one of R^3 and R^4 is hydrogen, and R^2 is fluoro, then the other of R^3 and R^4 is not hydrogen, halogen, azido, trifluoromethyl, C_{1-4} alkyl, amino, C_{1-4} alkylamino, alkyl)amino, or C_{1-10} alkoxy; (b) when R^1 is hydrogen, one of R^3 and R^4 is hydrogen, and R^2 is halogen, hydroxy, C_{1-6} alkoxy, or C_{2-6} alkenyloxy, then the other of R^3 and R^4 is not hydrogen, fluoro, or azido; and (c) when R^1 and R^3 are hydrogen and R^2 is hydroxy, then R^4 is not hydroxy.

In one embodiment of the present invention is the method of inhibiting RNA-dependent RNA viral polymerase, inhibiting RNA-dependent viral replication, and/or treating RNA-dependent RNA viral infection with a compound of structural formula II which is of the stereochemical configuration:

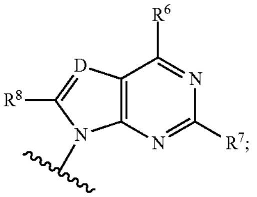


(II)

wherein B is



or



D is N, CH, C—CN, C—NO_2, C—C_{1-3} alkyl, C—NHCONH$_2$, C—CONR^{11}R^{11}, C—CSNR^{11}R^{11}, C—COOR^{11}, C-hydroxy, C—C_{1-3} alkoxy, C-amino, C—C_{1-4} alkylamino, C-di(C_{1-4} alkyl)amino, C-halogen, C-(1,3-oxazol-2-yl), C-(1,3-thiazol-2-yl), or C-(imidazol-2-yl);

wherein alkyl is unsubstituted or substituted with one to three groups independently selected from halogen, amino, hydroxy, carboxy, and

C_{1-3} alkoxy;

E is N or C—R^5;

W is O or S;

Y is H, C_{1-10} alkylcarbonyl, $P_3O_9H_4$, or $P(O)R^9R^{10}$;

R^1 is hydrogen, CF_3, or C_{1-4} alkyl and one of R^2 and R^3 is OH or C_{1-4} alkoxy and the other of

R^2 and R^3 is selected from the group consisting of
hydrogen,
hydroxy,
halogen,
C_{1-3} alkyl,
trifluoromethyl,
C_{1-4} alkoxy,
C_{1-4} alkylthio,
C_{1-8} alkylcarbonyloxy,
aryloxycarbonyl,
azido,
amino,
C_{1-4} alkylamino, and
di(C_{1-4} alkyl)amino; or

R^2 is hydrogen, CF_3, or C_{1-4} alkyl and one of R^1 and R^3 is OH or C_{1-4} alkoxy and the other of R^1 and R^3 is selected from the group consisting of
hydrogen,
hydroxy,
fluoro,
C_{1-4} alkyl,
trifluoromethyl,
C_{1-4} alkoxy,
C_{1-4} alkylthio,
alkylcarbonyloxy,
azido,
amino,
C_{1-4} alkylamino, and
di(C_{1-4} alkyl)amino; or

R^1 and R^2 together with the carbon atom to which they are attached form a 3- to 6-membered saturated monocyclic ring system optionally containing a heteroatom selected from O, S, and NC_{0-4} alkyl;

R^4 and R^6 are each independently H, OH, SH, NH_2, C_{1-4} alkylamino, alkyl)amino, C_{3-6} cycloalkylamino, halogen, C_{1-4} alkyl, C_{1-4} alkoxy, or CF_3;

R^5 is H, C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-4} alkylamino, CF_3, or halogen;

R^7 is hydrogen, amino, C_{1-4} alkylamino, C_{3-6} cycloalkylamino, or di(C_{1-4} alkyl)amino;

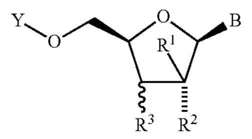
each R^{11} is independently H or C_{1-6} alkyl;

R^8 is H, halogen, CN, carboxy, C_{1-4} alkyloxycarbonyl, N_3, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, hydroxy, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} alkylsulfonyl, or (C_{1-4} alkyl)$_{0-2}$ aminomethyl; and

R^9 and R^{10} are each independently hydroxy, $OCH_2CH_2SC(=O)C_{1-4}$ alkyl, or $OCH_2O(C=O)C_{1-4}$ alkyl;

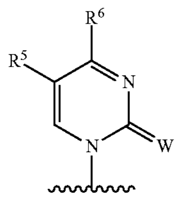
with the provisos that (a) when R^1 is hydrogen, one of R^3 and R^4 is hydrogen, and R^2 is fluoro, then the other of R^3 and R^4 is not hydrogen, halogen, trifluoromethyl, C_{1-4} alkyl, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, or C_{1-4} alkoxy; (b) when R^1 is hydrogen, one of R^3 and R^4 is hydrogen, and R^2 is halogen, hydroxy, or C_{1-4} alkoxy, then the other of R^3 and R^4 is not hydrogen, fluoro, or azido; and (c) when R^1 and R^3 are hydrogen and R^2 is hydroxy, then R^4 is not hydroxy.

In a second embodiment of the present invention is the method of inhibiting RNA-dependent RNA viral polymerase, inhibiting RNA-dependent RNA viral replication, and/or treating RNA-dependent RNA viral infection with a compound of structural formula III which is of the stereochemical configuration:

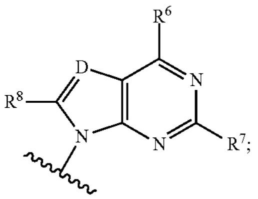


(III)

wherein B is



or



D is N, CH, C—CN, C—NO_2, C—C_{1-3} alkyl, C—NHCONH$_2$, C—CONR^{11}R^{11},

C—CSNR^{11}R^{11}, C—COOR^{11}, C-hydroxy, C—C_{1-3} alkoxy, C-amino, C—C_{1-4} alkylamino, C-di(C_{1-4} alkyl)amino, C-halogen, C-(1,3-oxazol-2-yl), C-(1,3-thiazol-2-yl), or C-(imidazol-2-yl); wherein alkyl is unsubstituted or substituted with one to three groups independently selected from halogen, amino, hydroxy, carboxy, and

C_{1-3} alkoxy;

W is O or S;

Y is H, C_{1-10} alkylcarbonyl, $P_3O_9H_4$, $P_2O_6H_3$, or P(O)R^9R^{10};

R^1 is hydrogen, CF_3, or C_{1-4} alkyl and one of R^2 and R^3 is OH or C_{1-4} alkoxy and the other of R^2 and R^3 is selected from the group consisting of

hydrogen,
hydroxy,
fluoro,
C_{1-3} alkyl,
trifluoromethyl,
C_{1-8} alkylcarbonyloxy,
C_{1-3} alkoxy, and
amino; or

R^2 is hydrogen, CF_3, or C_{1-4} alkyl and one of R^1 and R^3 is OH or C_{1-4} alkoxy and the other of R^1 and R^3 is selected from the group consisting of

hydrogen,
hydroxy,
fluoro,
C_{1-3} alkyl,
trifluoromethyl,
C_{1-8} alkylcarbonyloxy,
C_{1-3} alkoxy, and
amino; or

R^1 and R^2 together with the carbon atom to which they are attached form a 3- to 6-membered saturated monocyclic ring system optionally containing a heteroatom selected from O, S, and NC_{0-4} alkyl;

R^6 is H, OH, SH, NH_2, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, C_{3-6} cycloalkylamino, halogen, C_{1-4} alkyl, C_{1-4} alkoxy, or CF_3;

R^5 is H, C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-4} alkylamino, CF_3, or halogen;

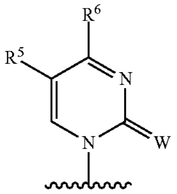
R^7 is hydrogen, amino, C_{1-4} alkylamino, C_{3-6} cycloalkylamino, or di(C_{1-4} alkyl)amino;

each R^{11} is independently H or C_{1-6} alkyl;

R^8 is H, halogen, CN, carboxy, C_{1-4} alkyloxycarbonyl, N_3, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, hydroxy, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} alkylsulfonyl, or (C_{1-4} alkyl)$_{0-2}$ aminomethyl; and

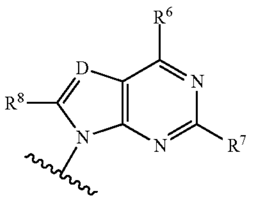
R^9 and R^{10} are each independently hydroxy, $OCH_2CH_2SC(=O)$t-butyl, or $OCH_2O(C=O)$iPr; with the provisos that (a) when R^1 is hydrogen and R^2 is fluoro, then R^3 is not hydrogen, trifluoromethyl, fluoro, C_{1-3} alkyl, amino, or C_{1-3} alkoxy; (b) when R^1 is hydrogen and R^2 is fluoro, hydroxy, or C_{1-3} alkoxy, then R^3 is not hydrogen or fluoro; and (c) when R^1 is hydrogen and R^2 is hydroxy, then R^3 is not β-hydroxy.

In a class of this embodiment is the method of inhibiting RNA-dependent RNA viral polymerase, inhibiting RNA-dependent RNA viral replication, and/or treating RNA-dependent RNA viral infection with a compound of structural formula III wherein B is



and W, Y, and the R substituents are as defined under this second embodiment.

In a second class of this embodiment is the method of inhibiting RNA-dependent RNA viral polymerase, inhibiting RNA-dependent RNA viral replication, and/or treating RNA-dependent RNA viral infection with a compound of structural formula III wherein B is



and Y, D, and the R substituents are as defined under this second embodiment.

In a third embodiment of the present invention, the RNA-dependent RNA viral polymerase is a positive-sense single-stranded RNA-dependent RNA viral polymerase. In a class of this embodiment, the positive-sense single-stranded RNA-dependent RNA viral polymerase is a Flaviviridae viral polymerase or a Picornaviridae viral polymerase. In a subclass of this class, the Picornaviridae viral polymerase is rhinovirus polymerase, poliovirus polymerase, or hepatitis A virus polymerase. In a second subclass of this class, the Flaviviridae viral polymerase is selected from the group consisting of hepatitis C virus polymerase, yellow fever virus polymerase, dengue virus polymerase, West Nile virus polymerase, Japanese encephalitis virus polymerase, Banzi virus polymerase, and bovine viral diarrhea virus (BVDV) polymerase. In a subclass of this subclass, the Flaviviridae viral polymerase is hepatitis C virus polymerase.

In a fourth embodiment of the present invention, the RNA-dependent RNA viral replication is a positive-sense single-stranded RNA-dependent RNA viral replication. In a class of this embodiment, the positive-sense single-stranded RNA-dependent RNA viral replication is Flaviviridae viral replication or Picornaviridae viral replication. In a subclass of this class, the Picornaviridae viral replication is rhinovirus replication, poliovirus replication, or hepatitis A virus replication. In a second subclass of this class, the Flaviviridae viral replication is selected from the group consisting of hepatitis C virus replication, yellow fever virus replication, dengue virus replication, West Nile virus replication, Japanese encephalitis virus replication, Banzi virus replication, and bovine viral diarrhea virus replication. In a subclass of this subclass, the Flaviviridae viral replication is hepatitis C virus replication.

In a fifth embodiment of the present invention, the RNA-dependent RNA viral infection is a positive-sense single-stranded RNA-dependent viral infection. In a class of this embodiment, the positive-sense single-stranded RNA-dependent RNA viral infection is Flaviviridae viral infection or Picornaviridae viral infection. In a subclass of this class, the Picornaviridae viral infection is rhinovirus infection, poliovirus infection, or hepatitis A virus infection. In a second subclass of this class, the Flaviviridae viral infection is selected from the group consisting of hepatitis C virus infection, yellow fever virus infection, dengue virus infection, West Nile virus infection, Japanese encephalitis virus infection, Banzi virus infection, and bovine viral diarrhea virus infection. In a subclass of this subclass, the Flaviviridae viral infection is hepatitis C virus infection.

Illustrative of the invention is a method for inhibiting RNA-dependent RNA viral polymerase, inhibiting RNA-dependent RNA viral replication, and/or treating RNA-dependent RNA viral infection wherein the compound is selected from:

2'-O-methyl-cytidine,
2'-C-methyl-cytidine,
3',5'-di-O-octanoyl-2'-O-methyl-cytidine,
3'-O-octanoyl-2'-O-methyl-cytidine,
2'-C-methyl-adenosine,
8-amino-2'-C-methyladenosine,
4-amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile,
4-amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carboxamide,
3'-deoxy-3'-methyl-cytidine,
4-amino-7-(3-deoxy-3-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide,
3'-deoxy-adenosine,
4-amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-c]pyrimidine,

4-amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-c]pyrimidin-5-carboxamide,
3'-amino-3'-deoxyadenosine,
2-amino-3,4-dihydro-4-oxo-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide,
4-amino-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide,
2-amino-3,4-dihydro-4-oxo-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-c]pyrimidin-5-carbonitrile,
2-amino-5-ethyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
6-amino-1-(β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one,
3'-deoxyguanosine,
2-amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2'-O-methylguanosine,
2-amino-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-7-(2-O-methyl-β-D-ribofuranosyl)-5H-pyrrolo[3,2-d]pyrimidin-4(3H)-one,
7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
3'-deoxycytidine,
2-amino-5-methyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-3,4-dihydro-4-oxo-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo-[2,3-d]pyrimidin-5-carbonitrile,
2-amino-5-methyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
8-azidoguanosine,
8-aminoguanosine,
8-bromoadenosine,
8-aminoadenosine,
8-bromoguanosine,
3'-deoxy-3'-fluorocytidine,
3'-deoxy-3'-fluoroguanosine,
4-amino-7-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide,
2-amino-4-chloro-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile,
2-amino-4-chloro-5-ethyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-4-chloro-5-methyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo-[2,3-d]pyrimidine,
2-amino-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-thione,
2-amino-4-chloro-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-c]pyrimidine,
2-amino-4-chloro-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-4-chloro-5-methyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
1-(β-D-ribofuranosyl)-1H-pyrazolo[3,4-d]pyrimidin-4(3H)-one,
4-amino-1-(β-D-ribofuranosyl)-1H-pyrazolo[3,4-d]pyrimidine,
2-amino-6-chloro-9-(β-D-ribofuranosyl)-9H-purine,
2-amino-4-chloro-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile,
6-methyl-9-(β-D-ribofuranosyl)-9H-purine,
2-amino-7-(2-deoxy-2-fluoro-β-D-arabino furanosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-4-chloro-7-(2-deoxy-2-fluoro-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,

2-amino-7-(β-D-arabinofuranosyl)-7H-pyrrolo[2,3-c]pyrimidin-4(3H)-one,
2-amino-7-(β-D-arabinofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile,
2-amino-5-methyl-7-(β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
9-(β-D-arabino furanosyl)-9H-purin-6(1H)-one,
1-(β-D-arabinofuranosyl)-1H-cytosine,
2-amino-4-chloro-5-methyl-7-(β-D-arabinofuranosyl)-7H-pyrrolo[2,3-c]pyrimidine,
3'-deoxy-3'-(fluoromethyl)-guanosine,
2'-amino-2'-deoxycytidine,
4-amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-c]pyrimidin-5-carbonitrile,
2'-O-methyladenosine,
4-amino-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
3'-amino-3'-deoxy-2'-O-methyl-adenosine,
3'-deoxy-3'-methyl-uridine,
6-amino-1-(3-deoxy-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one,
6-amino-1-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(3H)-one,
3'-deoxy-3'-fluorouridine,
3'-deoxy-3'-fluoroadenosine,
2-amino-7-(2-deoxy-β-D-ribofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo pyrimidin-5-carbonitrile,
3'-deoxy-5-methyl-uridine,
3'-deoxy-2'-O-(2-methoxyethyl)-3'-methyl-5-methyluridine,
2'-amino-2'-deoxy-uridine,
2-amino-9-(β-D-arabinofuranosyl)-9H-purin-6(1H)-one,
3'-deoxy-3'-methylguanosine,
2'-O-[4-(imidazolyl-1)butyl]guanosine,
2'-deoxy-2'-fluoroguanosine,
2'-deoxyguanosine,
2-amino-7-(2-deoxy-2-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-c]pyrimidin-4(3H)-one,
2-amino-7-(3-deoxy-β-D-ribofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile,
2-amino-5-iodo-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-7-(3-deoxy-3-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile,
2-amino-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-7-(2-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-7-(3-deoxy-3-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-7-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-c]pyrimidin-4(3H)-one,
6-amino-1-(2-O-methyl-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one,
6-amino-1-(2-deoxy-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one,
6-amino-1-(3-deoxy-3-methyl-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one,
6-amino-1-(2-deoxy-2-fluoro-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one,
6-amino-1-(β-D-arabinofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one,
2'-O-[2-(N,N-diethylaminooxy)ethyl]-5-methyluridine,
5-ethynyl-2'-O-(2-methoxyethyl)-cytidine,
1-(2-C-methyl-β-D-arabinofuranosyl)uracil,
5-methyl-3'-deoxycytidine,
2-amino-2'-O-methyladenosine,
2'-deoxy-2'-fluoro adenosine,
3'-deoxy-3'-fluoroadenosine,
3'-deoxy-3'-methyladenosine,
2-amino-7-(2-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-7-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide,
4-amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide,
4-amino-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-7-(β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-1-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridine,
4-amino-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine (tubercidin),
4,6-diamino-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-7-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo-[2,3-d]-pyrimidin-5-carboxamide,
4-amino-1-(3-deoxy-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridine,
4-amino-1-(3-deoxy-3-methyl-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridine,
4-amino-1-(β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridine,
4-amino-1-(2-C-methyl-β-D-ribofuranosyl)-1H-pyrazolo[3,4-d]pyrimidine,
4-amino-7-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine; and
the corresponding 5'-triphosphates, 5'-[bis(isopropyloxycarbonyloxymethyl)]monophosphates, 5'-mono-(S—C_{1-4} alkanoyl-2-thioethyl)monophosphates, and 5'-bis-(S—C_{1-4} alkanoyl-2-thioethyl)monophosphates thereof;
or a pharmaceutically acceptable salt thereof.

Further illustrative of the invention is a method for inhibiting RNA-dependent RNA viral polymerase, inhibiting RNA-dependent RNA viral replication, and/or treating RNA-dependent RNA viral infection wherein the compound is selected from:

2'-O-methyl-cytidine,
2'-C-methyl-cytidine,
3',5'-di-O-octanoyl-2'-O-methyl-cytidine,
3'-O-octanoyl-2'-O-methyl-cytidine,
4-amino-1(θ-D-ribofuranosyl)-1H-pyrazolo[3,4-d]pyrimidine,
4-amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile,
4-amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carboxamide, 2'-C-methyladenosine,
8-amino-2'-C-methyladenosine,
3'-deoxy-3'-methyl-cytidine,
4-amino-7-(3-deoxy-3-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide,
3'-deoxyadenosine,
4-amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide,
3'-amino-3'-deoxyadenosine,
2-amino-3,4-dihydro-4-oxo-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide,
4-amino-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide,
2-amino-3,4-dihydro-4-oxo-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile,
2-amino-5-ethyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,

6-amino-1-(β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one,
3'-deoxyguanosine,
2-amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2'-O-methylguanosine,
2-amino-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-7-(2-O-methyl-β-D-ribofuranosyl)-5H-pyrrolo[3,2-d]pyrimidin-4-(3H)-one,
7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
3'-deoxy-cytidine,
2-amino-5-methyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-3,4-dihydro-4-oxo-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo-[2,3-d]pyrimidine-5-carbonitrile,
2-amino-5-methyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
8-azidoguanosine,
8-aminoguanosine,
8-bromoadenosine,
8-aminoadenosine,
8-bromoguanosine,
3'-deoxy-3'-fluorocytidine,
3'-deoxy-3'-fluoroguanosine,
4-amino-7-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide,
2-amino-4-chloro-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile,
2-amino-4-chloro-5-ethyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-4-chloro-5-methyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo-[2,3-c]pyrimidine,
2-amino-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-c]pyrimidine,
2-amino-4-chloro-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-4-chloro-5-methyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-7-(2-deoxy-2-fluoro-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
4-amino-1-(2-C-methyl-β-D-ribofuranosyl)-1H-pyrazolo[3,4-d]pyrimidine,
2-amino-7-(β-D-arabinofuranosyl)-7H-pyrrolo[2,3-c]pyrimidin-4(3H)-one, and
2-amino-7-(β-D-arabinofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile; and
the corresponding 5'-triphosphates, 5'-[bis(isopropyloxycarbonyloxymethyl)]monophosphates, 5'-mono-(S-pivaloyl-2-thioethyl)monophosphates, and 5'-bis-(S-pivaloyl-2-thioethyl)monophosphates thereof;
or a pharmaceutically acceptable salt thereof.

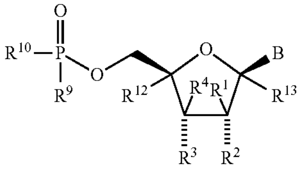
Even further illustrative of the present invention is a method for inhibiting RNA-dependent RNA viral polymerase, inhibiting RNA-dependent RNA viral replication, and/or treating RNA-dependent RNA viral infection wherein the compound is selected from
2'-O-methyl-cytidine,
2'-C-methyl-cytidine,
3',5'-di-O-octanoyl-2'-O-methyl-cytidine,
3'-O-octanoyl-2'-O-methyl-cytidine,
4-amino-1-(9-D-ribofuranosyl)-1H-pyrazolo[3,4-d]pyrimidine,
4-amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile,

4-amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carboxamide,
2'-C-methyladenosine,
8-amino-2'-C-methyladenosine,
8-bromoguanosine,
8-aminoguanosine,
8-aminoadenosine,
4-amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-4-chloro-5-ethyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-3,4-dihydro-4-oxo-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide,
4-amino-1-(2-C-methyl-β-D-ribofuranosyl)-1H-pyrazolo[3,4-d]pyrimidine,
2-amino-4-chloro-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile;
and the corresponding 5'-triphosphates thereof;
2'-O-methylcytidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate],
2-amino-7-(3-deoxy-β-D-ribofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo[2,3-d]pyrimidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate],
3'-deoxyguanosine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate], and
3'-deoxycytidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate];
or a pharmaceutically acceptable salt thereof.

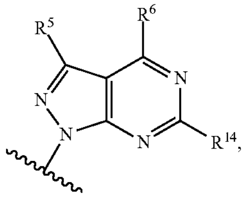
Yet further illustrative of the invention is a method for inhibiting RNA-dependent RNA viral polymerase, inhibiting RNA-dependent RNA viral replication, and/or treating RNA-dependent RNA viral infection wherein the compound is selected from:
2'-O-methylcytidine,
2'-C-methylcytidine,
3',5'-di-O-octanoyl-2'-O-methyl-cytidine,
3'-O-octanoyl-2'-O-methyl-cytidine,
4-amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile,
4-amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carboxamide,
2'-C-methyladenosine,
8-amino-2'-C-methyladenosine,
2'-O-methylcytidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate],
2-amino-7-(3-deoxy-β-D-ribofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo[2,3-d]pyrimidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate], and
3'-deoxycytidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate];
or a pharmaceutically acceptable salt thereof.

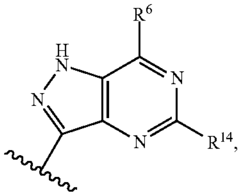
The present invention also provides novel compounds of structural formula IV of the indicated stereochemical configuration which are useful as inhibitors of RNA-dependent RNA viral polymerase:

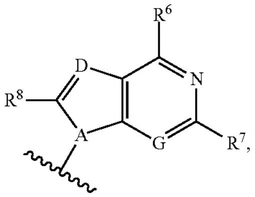
(IV)

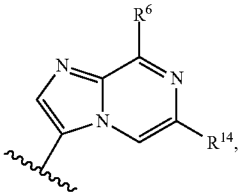


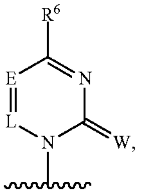
wherein B is selected from the group consisting of

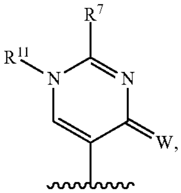




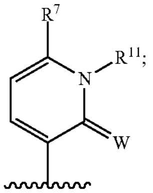








and



A, G, and L are each independently CH or N;

D is N, CH, C—CN, C—NO_2, C—C_{1-3} alkyl, C—NHCONH$_2$, C—CONR^{11}R^{11}, C—CSNR^{11}R^{11}, C—COOR^{11}, C-hydroxy, C—C_{1-3} alkoxy, C-amino, C—C_{1-4} alkylamino, C-di(C_{1-4} alkyl)amino, C-halogen, C-(1,3-oxazol-2-yl), C-(1,3-thiazol-2-yl), or C-(imidazol-2-yl); wherein alkyl is unsubstituted or substituted with one to three groups independently selected from halogen, amino, hydroxy, carboxy, and C_{1-3} alkoxy;

E is N or CRS;

W is O or S;

R^1 is hydrogen, C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms and one of R^2 and R^3 is hydroxy or C_{1-4} alkoxy and the other of R^2 and R^3 is selected from the group consisting of

- hydrogen,
- hydroxy,
- halogen,
- C_{1-4} alkyl, optionally substituted with 1 to 3 fluorine atoms,
- C_{1-10} alkoxy, optionally substituted with C_{1-3} alkoxy or 1 to 3 fluorine atoms,
- C_{2-6} alkenyloxy,
- C_{1-4} alkylthio,
- C_{1-8} alkylcarbonyloxy,
- aryloxycarbonyl,
- azido,
- amino,
- C_{1-4} alkylamino, and
- di(C_{1-4} alkyl)amino; or

R^2 is hydrogen, C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms and one of R^1 and R^3 is hydroxy or C_{1-4} alkoxy and the other of R^1 and R^3 is selected from the group consisting of

- hydrogen,
- hydroxy,
- halogen,
- C_{1-4} alkyl, optionally substituted with 1 to 3 fluorine atoms,
- C_{1-10} alkoxy, optionally substituted with hydroxy, C_{1-3} alkoxy, carboxy, or 1 to 3 fluorine atoms,
- C_{2-6} alkenyloxy,
- C_{1-4} alkylthio,
- C_{1-8} alkylcarbonyloxy,
- aryloxycarbonyl,
- azido,
- amino,
- C_{1-4} alkylamino, and
- di(C_{1-4} alkyl)amino; or

R^1 and R^2 together with the carbon atom to which they are attached form a 3- to 6-membered saturated monocyclic ring system optionally containing a heteroatom selected from O, S, and NC_{0-4} alkyl;

R^4 and R^6 are each independently H, OH, SH, NH_2, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, C_{3-6} cycloalkylamino, halogen, C_{1-4} alkyl, C_{1-4} alkoxy, or CF_3;

R^5 is H, C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-4} alkylamino, CF_3, or halogen;

R^{14} is H, CF_3, C_{1-4} alkyl, amino, C_{1-4} alkylamino, C_{3-6} cycloalkylamino, or di(C_{1-4} alkyl)amino;

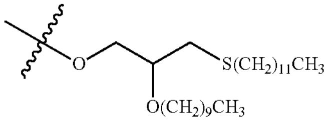
R^7 is hydrogen, amino, C_{1-4} alkylamino, C_{3-6} cycloalkylamino, or di(C_{1-4} alkyl)amino;

each R^{11} is independently H or C_{1-6} alkyl;

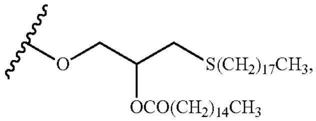
R^8 is H, halogen, CN, carboxy, C_{1-4} alkyloxycarbonyl, N_3, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, hydroxy, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} alkylsulfonyl, or (C_{1-4} alkyl)$_{0-2}$ aminomethyl;

R^{12} and R^{13} are each independently hydrogen or methyl; and

R^9 and R^{10} are each independently hydroxy, $OCH_2CH_2SC({=}O)C_{1-4}$ alkyl, $OCH_2O(C{=}O)OC_{1-4}$ alkyl, $NHCHMeCO_2Me$, $OCH(C_{1-4}$ alkyl$)O(C{=}O)C_{1-4}$ alkyl,



or

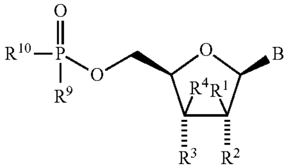


provided that at least one of R^9 and R^{10} is not hydroxy.

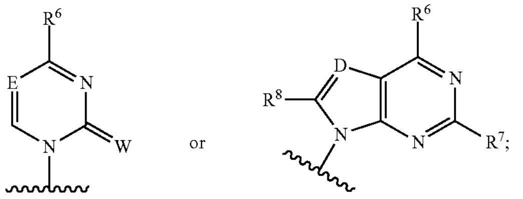
The compounds of formula IV are also inhibitors of RNA-dependent RNA viral replication and are useful for the treatment of RNA-dependent RNA viral infection.

In one embodiment, there are provided novel compounds of structural formula V which are of the stereochemical configuration:

(V)



wherein B is



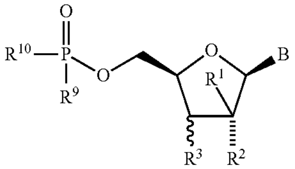
D is N, CH, C—CN, C—NO_2, C—C_{1-3} alkyl, C—NHCONH$_2$, C—CONR^{11}R^{11},
C—CSNR^{11}R^{11}, C—COOR^{11}, C-hydroxy, C—C_{1-3} alkoxy, C-amino, C—C_{1-4} alkylamino, C-di(C_{1-4} alkyl)amino, C-halogen, C-(1,3-oxazol-2-yl), C-(1,3-thiazol-2-yl), or C-(imidazol-2-yl);
wherein alkyl is unsubstituted or substituted with one to three groups independently selected from halogen, amino, hydroxy, carboxy, and
C_{1-3} alkoxy;
W is O or S;
E is N or C—R^5;
R^1 is hydrogen, C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms and one of R^2 and R^3 is hydroxy or C_{1-4} alkoxy and the other of R^2 and R^3 is selected from the group consisting of
- hydrogen,
- hydroxy,
- halogen,
- C_{1-3} alkyl,
- trifluoromethyl,
- C_{1-4} alkoxy,
- C_{1-4} alkylthio,
- C_{1-8} alkylcarbonyloxy,
- aryloxycarbonyl,
- azido,
- amino,
- C_{1-4} alkylamino, and
- di(C_{1-4} alkyl)amino; or

R^2 is hydrogen, C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms and one of R^1 and R^3 is hydroxy or C_{1-4} alkoxy and the other of R^1 and R^3 is selected from the group consisting of
- hydrogen,
- hydroxy,
- fluoro,
- C_{1-4} alkyl,
- trifluoromethyl,
- C_{1-4} alkoxy,
- C_{1-4} alkylthio,
- C_{1-8} alkylcarbonyloxy,
- azido,
- amino,
- C_{1-4} alkylamino, and
- di(C_{1-4} alkyl)amino; or

R^1 and R^2 together with the carbon atom to which they are attached form a 3- to 6-membered saturated monocyclic ring system optionally containing a heteroatom selected from O, S, and NC_{0-4} alkyl;
R^4 and R^6 are each independently H, OH, SH, NH_2, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, C_{3-6} cycloalkylamino, halogen, C_{1-4} alkyl, C_{1-4} alkoxy, or CF_3;
R^5 is H, C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-4} alkylamino, CF_3, or halogen;

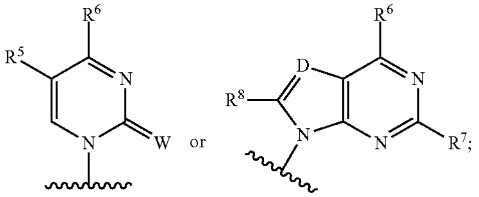
R^7 is hydrogen, amino, C_{1-4} alkylamino, C_{3-6} cycloalkylamino, or di(C_{1-4} alkyl)amino;
each R^{11} is independently H or C_{1-6} alkyl;
R^8 is H, halogen, CN, carboxy, C_{1-4} alkyloxycarbonyl, N_3, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, hydroxy, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} alkylsulfonyl, or (C_{1-4} alkyl)$_{0-2}$ aminomethyl; and
R^9 and R^{10} are each independently hydroxy, OCH_2CH_2SC(═O)C_{1-4} alkyl, or OCH_2O(C═O)C_{1-4} alkyl, provided that at least one of R^9 and R^{10} is not hydroxy.

In a second embodiment, there are provided novel compounds of structural formula VI:



(VI)

wherein B is



D is N, CH, C—CN, C—NO_2, C—C_{1-3} alkyl, C—NHCONH$_2$, C—CONR^{11}R^{11},
C—CSNR^{11}R^{11}, C—COOR^{11}, C-hydroxy, C—C_{1-3} alkoxy, C-amino, C—C_{1-4} alkylamino, C-di(C_{1-4} alkyl)amino, C-halogen, C-(1,3-oxazol-2-yl), C-(1,3-thiazol-2-yl), or C-(imidazol-2-yl); wherein alkyl is unsubstituted or substituted with one to three groups independently selected from halogen, amino, hydroxy, carboxy, and
C_{1-3} alkoxy;
W is O or S;
R^1 is hydrogen, C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms and one of R^2 and R^3 is hydroxy or C_{1-4} alkoxy and the other of R^2 and R^3 is selected from the group consisting of
- hydrogen,
- hydroxy,
- fluoro,
- C_{1-3} alkyl,
- trifluoromethyl,
- C_{1-3} alkoxy,
- C_{1-8} alkylcarbonyloxy, and
- amino; or

R^2 is hydrogen, C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms and one of R^1 and R^3 is hydroxy or C_{1-4} alkoxy and the other of R^1 and R^3 is selected from the group consisting of
- hydrogen,
- hydroxy,
- fluoro,
- C_{1-3} alkyl,

trifluoromethyl,

C_{1-3} alkoxy,

C_{1-8} alkylcarbonyloxy, and

amino; or

R^1 and R^2 together with the carbon atom to which they are attached form a 3- to 6-membered saturated monocyclic ring system optionally containing a heteroatom selected from O, S, and NC_{0-4} alkyl;

R^6 is H, OH, SH, NH_2, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, C_{3-6} cycloalkylamino, halogen, C_{1-4} alkyl, C_{1-4} alkoxy, or CF_3;

R^5 is H, C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-4} alkylamino, CF_3, or halogen;

R^7 is hydrogen, amino, C_{1-4} alkylamino, C_{3-6} cycloalkylamino, or di(C_{1-4} alkyl)amino;

each R^{11} is independently H or C_{1-6} alkyl;

R^8 is H, halogen, CN, carboxy, C_{1-4} alkyloxycarbonyl, N_3, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, hydroxy, C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} alkylsulfonyl, or (C_{1-4} alkyl)$_{0-2}$ aminomethyl; and

R^9 and R^{10} are each independently hydroxy, $OCH_2CH_2SC({=}O)$t-butyl, or $OCH_2O(C{=}O)$iPr, provided that at least one of R^9 and R^{10} is not hydroxy.

Illustrative of the novel compounds of structural formula VI of the present invention are the following:

2'-O-methylcytidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate],

2-amino-7-(3-deoxy-β-D-ribofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo[2,3-d]pyrimidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate],

3'-deoxyguanosine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate],

2'-O-methylguanosine-5'-[bis-(S-acetyl-2-thioethyl)phosphate],

2'-O-methylguanosine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate],

8-bromo-2'-O-methylguanosine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate],

2-amino-3,4-dihydro-7-(2-O-methyl-β-D-ribofuranosyl)-4-oxo-7H-pyrrolo[2,3-d]pyrimidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate],

2-amino-5-bromo-7-(3-deoxy-β-D-ribofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo[2,3-d]pyrimidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate],

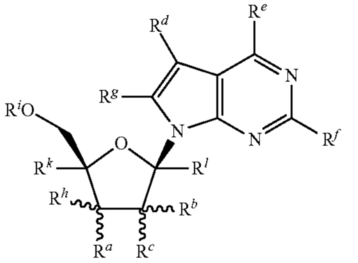
5-bromo-2'-O-methylcytidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate],

3'-deoxycytidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate], and

2'-O-methylcytidine-5'-[bis(isopropyloxycarbonyloxymethyl)]phosphate.

The present invention further provides novel compounds of structural formula XII of the indicated stereochemical configuration or a pharmaceutically acceptable salt thereof which are useful as inhibitors of RNA-dependent RNA viral polymerase:

(XII)



wherein R^a and R^h are each independently selected from the group consisting of hydrogen, cyano, azido, halogen, hydroxy, mercapto, amino, C_{1-4} alkoxy, C_{2-4} alkenyl, C_{2-4} alkynyl, and C_{1-4} alkyl, wherein alkyl is unsubstituted or substituted with hydroxy, amino, C_{1-4} alkoxy, C_{1-4} alkylthio, or one to three fluorine atoms;

R^b is C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl, wherein alkyl is unsubstituted or substituted with hydroxy, amino, C_{1-4} alkoxy, C_{1-4} alkylthio, or one to three fluorine atoms;

R^c is hydrogen, fluorine, hydroxy, mercapto, C_{1-4} alkoxy, or C_{1-4} alkyl; or R^b and R^c together with the carbon atom to which they are attached form a 3- to 6-membered saturated monocyclic ring system optionally containing a heteroatom selected from O, S, and NC_{0-4} alkyl;

R^d is hydrogen, cyano, nitro, C_{1-3} alkyl, $NHCONH_2$, CONRjRj, CSNRjRj, COORj, $C({=}NH)NH_2$, hydroxy, C_{1-3} alkoxy, amino, C_{1-4} alkylamino, alkyl)amino, halogen, (1,3-oxazol-2-yl), (1,3-thiazol-2-yl), or (imidazol-2-yl); wherein alkyl is unsubstituted or substituted with one to three groups independently selected from halogen, amino, hydroxy, carboxy, and C_{1-3} alkoxy;

R^e and R^f are each independently hydrogen, hydroxy, halogen, C_{1-4} alkoxy, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, C_{3-6} cycloalkylamino, di(C_{3-6} cycloalkyl)amino, or C_{4-6} cycloheteroalkyl, unsubstituted or substituted with one to two groups independently selected from halogen, hydroxy, amino, C_{1-4} alkyl, and

C_{1-4} alkoxy;

R^g is hydrogen, C_{1-4} alkyl, C_{2-4} alkynyl, halogen, cyano, carboxy, C_{1-4} alkyloxycarbonyl, azido, amino, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, hydroxy,

C_{1-6} alkoxy, C_{1-6} alkylthio, C_{1-6} alkylsulfonyl, (C_{1-4} alkyl)$_{0-2}$ aminomethyl, or

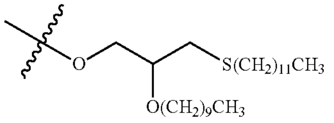
C_{4-6} cycloheteroalkyl, unsubstituted or substituted with one to two groups independently selected from halogen, hydroxy, amino, C_{1-4} alkyl, and C_{1-4} alkoxy;

R^i is hydrogen, C_{1-10} alkylcarbonyl, $P_3O_9H_4$, $P_2O_6H_3$, or $P(O)R^mR^n$;

each R^j is independently hydrogen or C_{1-6} alkyl;

R^k and R^l are each independently hydrogen, methyl, hydroxymethyl, or fluoromethyl; and

R^m and R^n are each independently hydroxy, $OCH_2CH_2SC({=}O)C_{1-4}$ alkyl, $OCH_2O(C{=}O)OC_{1-4}$ alkyl, $NHCHMeCO_2Me$, $OCH(C_{1-4}\ alkyl)O(C{=}O)C_{1-4}$ alkyl,



or

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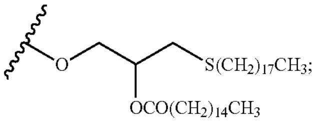
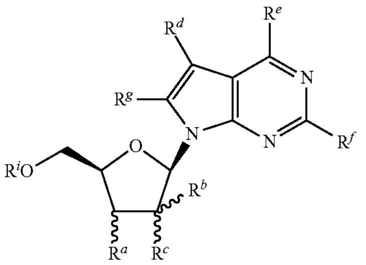


with the proviso that when R^a and R^c are α-hydroxy, R^e is amino, R^b is β-methyl and R^h is hydrogen or R^h is β-methyl and R^b is hydrogen, and R^f, R^g, R^i, R^k, and R^l are hydrogen, then R^d is not cyano or $CONH_2$.

The compounds of formula XII are also inhibitors of RNA-dependent RNA viral replication and are useful for the treatment of RNA-dependent RNA viral infection.

In one embodiment of the novel compounds of structural formula XII are the compounds of structural formula XIII:



XIII

wherein R^a is hydrogen, halogen, hydroxy, amino, or C_{1-3} alkoxy;

R^b is C_{1-3} alkyl, wherein alkyl is unsubstituted or substituted with hydroxy, amino, C_{1-3} alkoxy, C_{1-3} alkylthio, or one to three fluorine atoms;

R^c is hydroxy, fluoro, or C_{1-4} alkoxy;

R^d is hydrogen, cyano, methyl, halogen, or $CONH_2$;

R^g is hydrogen, amino, or C_{1-4} alkylamino;

R^i is hydrogen, $P_3O_9H_4$, $P_2O_6H_3$, or PO_3H_2; and

R^e and R^f are each independently hydrogen, hydroxy, halogen, amino,

C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, or C_{3-6} cycloalkylamino;

with the proviso that when R^a and R^c are α-hydroxy, R^e is amino, R^b is methyl, and R^f, R^g, and

R^i are hydrogen, then R^d is not cyano or $CONH_2$.

In a second embodiment of the compounds of structural formula XII are the compounds of structural formula XIII wherein:

R^b is methyl, fluoromethyl, hydroxymethyl, difluoromethyl, trifluoromethyl, or aminomethyl;

R^c is hydroxy, fluoro, or methoxy;

R^a is hydrogen, fluoro, hydroxy, amino, or methoxy;

R^i is hydrogen or $P_3O_9H_4$;

R^g is hydrogen or amino;

R^d is hydrogen, cyano, methyl, halogen, or $CONH_2$; and

R^e and R^f are each independently hydrogen, fluoro, hydroxy, or amino;

with the proviso that when R^b is β-methyl, R^a and R^c are α-hydroxy, R^e is amino, and R^f, R^g, and R^i are hydrogen, then R^d is not cyano or $CONH_2$.

Illustrative of the novel compounds of the present invention of structural formula XIII which are useful as inhibitors of RNA-dependent RNA viral polymerase are the following:

4-amino-7-(2-C-methyl-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,

4-amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-c]pyrimidine,

4-methylamino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,

4-dimethylamino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,

4-cyclopropylamino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,

4-amino-7-(2-C-vinyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,

4-amino-7-(2-C-hydroxymethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,

4-amino-7-(2-C-fluoromethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,

4-amino-5-methyl-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,

4-amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carboxylic acid,

4-amino-5-bromo-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,

4-amino-5-chloro-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,

4-amino-5-fluoro-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-c]pyrimidine,

2,4-diamino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,

2-amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-c]pyrimidine,

2-amino-4-cyclopropylamino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,

2-amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,

4-amino-7-(2-C-ethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,

4-amino-7-(2-C,2-O-dimethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,

7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,

2-amino-5-methyl-7-(2-C,2-O-dimethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,

4-amino-7-(3-deoxy-2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,

4-amino-7-(3-deoxy-2-C-methyl-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,

4-amino-2-fluoro-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,

4-amino-7-(3-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,

4-amino-7-(3-C-methyl-β-D-xylofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,

4-amino-7-(2,4-di-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine, and

4-amino-7-(3-deoxy-3-fluoro-2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine;

and the corresponding 5'-triphosphates;

or a pharmaceutically acceptable salt thereof.

Further illustrative of the novel compounds of the present invention of structural formula XIII which are useful as inhibitors of RNA-dependent RNA viral polymerase are the following:

4-amino-7-(2-C-methyl-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,

4-amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,

4-amino-7-(2-C-fluoromethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,

4-amino-5-methyl-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-5-bromo-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-5-chloro-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
4-amino-5-fluoro-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine, and
4-amino-7-(2-C,2-O-dimethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
and the corresponding 5'-triphosphates;
or a pharmaceutically acceptable salt thereof.

Further structurally novel nucleoside derivatives of the present invention which are useful as inhibitors of RNA-dependent RNA viral polymerase are the following:

3'-deoxy-3'-methyl-cytidine,
3',5'-di-O-octanoyl-2'-O-methyl-cytidine,
3'-O-octanoyl-2'-O-methyl-cytidine,
4-amino-7-(3-deoxy-3-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide,
2-amino-5-ethyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-7-(2-O-methyl-β-D-ribofuranosyl)-5H-pyrrolo[3,2-d]pyrimidin-4(3H)-one,
7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-3,4-dihydro-4-oxo-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo-[2,3-d]-pyrimidin-5-carbonitrile,
2-amino-5-methyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-4-chloro-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile,
2-amino-4-chloro-5-ethyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-4-chloro-5-methyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-thione,
2-amino-4-chloro-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-4-chloro-5-methyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-7-(2-deoxy-2-fluoro-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-4-chloro-7-(2-deoxy-2-fluoro-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine,
2-amino-7-(β-D-arabinofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile,
9-(β-D-arabinofuranosyl)-9H-purin-6(1H)-one,
3'-amino-3'-deoxy-2'-O-methyl-adenosine,
8-amino-2'-C-methyladenosine,
6-amino-1-(3-deoxy-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one,
6-amino-1-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(3H)-one,
3'-deoxy-2'-O-(2-methoxyethyl)-3'-methyl-5-methyluridine,
2-amino-7-(3-deoxy-β-D-ribofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo[2,3-c]pyrimidin-5-carbonitrile,
2-amino-7-(3-deoxy-3-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile,
2-amino-7-(3-deoxy-3-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
2-amino-7-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one,
6-amino-1-(2-O-methyl-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one,
6-amino-1-(3-deoxy-3-methyl-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one,
6-amino-1-(2-deoxy-2-fluoro-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one,
1-(2-C-methyl-β-D-arabinofuranosyl)uracil,
4-amino-1-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridine,
2-amino-7-(–3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo-[2,3-d]pyrimidin-5-carboxamide,
4-amino-1-(2-C-methyl-β-D-ribofuranosyl)-1H-pyrazolo[3,4-d]pyrimidine,
4-amino-1-(3-deoxy-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridine, and
4-amino-1-(3-deoxy-3-methyl-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridine;
and the corresponding 5'-triphosphates;
or a pharmaceutically acceptable salt thereof.

In a further embodiment the novel compounds of the present invention are useful as inhibitors of positive-sense single-stranded RNA-dependent RNA viral polymerase, inhibitors of positive-sense single-stranded RNA-dependent RNA viral replication, and/or for the treatment of positive-sense single-stranded RNA-dependent RNA viral infection. In a class of this embodiment, the positive-sense single-stranded RNA-dependent RNA virus is a Flaviviridae virus or a Picornaviridae virus. In a subclass of this class, the Picornaviridae virus is a rhinovirus, a poliovirus, or a hepatitis A virus. In a second subclass of this class, the Flaviviridae virus is selected from the group consisting of hepatitis C virus, yellow fever virus, dengue virus, West Nile virus, Japanese encephalitis virus, Banzi virus, and bovine viral diarrhea virus (BVDV). In a subclass of this subclass, the Flaviviridae virus is hepatitis C virus.

Throughout the instant application, the following terms have the indicated meanings:

The alkyl groups specified above are intended to include those alkyl groups of the designated length in either a straight or branched configuration. Exemplary of such alkyl groups are methyl, ethyl, propyl, isopropyl, butyl, sec-butyl, tertiary butyl, pentyl, isopentyl, hexyl, isohexyl, and the like.

The term "alkenyl" shall mean straight or branched chain alkenes of two to six total carbon atoms, or any number within this range (e.g., ethenyl, propenyl, butenyl, pentenyl, etc.).

The term "alkynyl" shall mean straight or branched chain alkynes of two to six total carbon atoms, or any number within this range (e.g., ethynyl, propynyl, butynyl, pentynyl, etc.).

The term "cycloalkyl" shall mean cyclic rings of alkanes of three to eight total carbon atoms, or any number within this range (i.e., cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, cycloheptyl, or cyclooctyl).

The term "cycloheteroalkyl" is intended to include non-aromatic heterocycles containing one or two heteroatoms selected from nitrogen, oxygen and sulfur. Examples of 4-6-membered cycloheteroalkyl include azetidinyl, pyrrolidinyl, piperidinyl, morpholinyl, thiamorpholinyl, imidazolidinyl, tetrahydrofuranyl, tetrahydropyranyl, tetrahydrothiophenyl, piperazinyl, and the like.

The term "alkoxy" refers to straight or branched chain alkoxides of the number of carbon atoms specified (e.g., C_{1-4} alkoxy), or any number within this range [i.e., methoxy (MeO—), ethoxy, isopropoxy, etc.].

The term "alkylthio" refers to straight or branched chain alkylsulfides of the number of carbon atoms specified (e.g.,

C_{1-4} alkylthio), or any number within this range [i.e., methylthio (MeS—), ethylthio, isopropylthio, etc.].

The term "alkylamino" refers to straight or branched alkylamines of the number of carbon atoms specified (e.g., C_{1-4} alkylamino), or any number within this range [i.e., methylamino, ethylamino, isopropylamino, t-butylamino, etc.].

The term "alkylsulfonyl" refers to straight or branched chain alkylsulfones of the number of carbon atoms specified (e.g., C_{1-6} alkylsulfonyl), or any number within this range [i.e., methylsulfonyl ($MeSO_2$—), ethylsulfonyl, isopropylsulfonyl, etc.].

The term "alkyloxycarbonyl" refers to straight or branched chain esters of a carboxylic acid derivative of the present invention of the number of carbon atoms specified (e.g., C_{1-4} alkyloxycarbonyl), or any number within this range [i.e., methyloxycarbonyl (MeOCO—), ethyloxycarbonyl, or butyloxycarbonyl].

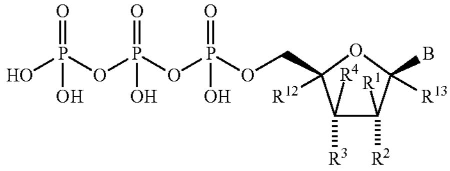
The term "aryl" includes both phenyl, naphthyl, and pyridyl. The aryl group is optionally substituted with one to three groups independently selected from C_{1-4} alkyl, halogen, cyano, nitro, trifluoromethyl, C_{1-4} alkoxy, and C_{1-4} alkylthio.

The term "halogen" is intended to include the halogen atoms fluorine, chlorine, bromine and iodine.

The term "substituted" shall be deemed to include multiple degrees of substitution by a named substituent. Where multiple substituent moieties are disclosed or claimed, the substituted compound can be independently substituted by one or more of the disclosed or claimed substituent moieties, singly or plurally.

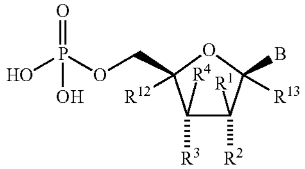
The term "5'-triphosphate" refers to a triphosphoric acid ester derivative of the 5'-hydroxyl group of a nucleoside compound of the present invention having the following general structural formula VII:

VII



wherein B, Z, R^1-R^4, R^{12}, and R^{13} are as defined above. The compounds of the present invention are also intended to include pharmaceutically acceptable salts of the triphosphate ester as well as pharmaceutically acceptable salts of 5'-monophosphate and 5'-diphosphate ester derivatives of the structural formulae VDT and IX, respectively,

VIII



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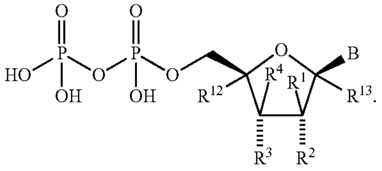
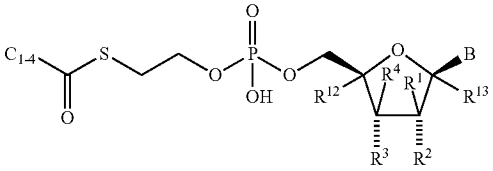
IX

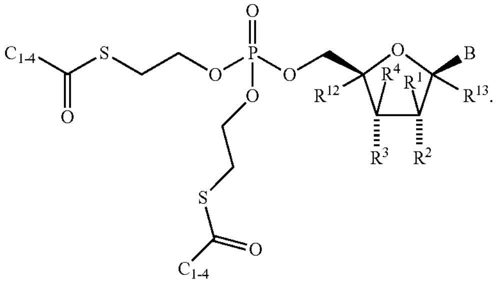


The term "5'-(S-acyl-2-thioethyl)phosphate" or "SATE" refers to a mono- or di-ester derivative of a 5'-monophosphate nucleoside of the present invention of structural formulae X and XI, respectively, as well as pharmaceutically acceptable salts of the mono-ester,

X



XI



The term "composition", as in "pharmaceutical composition," is intended to encompass a product comprising the active ingredient(s) and the inert ingredient(s) that make up the carrier, as well as any product which results, directly or indirectly, from combination, complexation or aggregation of any two or more of the ingredients, or from dissociation of one or more of the ingredients, or from other types of reactions or interactions of one or more of the ingredients. Accordingly, the pharmaceutical compositions of the present invention encompass any composition made by admixing a compound of the present invention and a pharmaceutically acceptable carrier.

The terms "administration of" and "administering a" compound should be understood to mean providing a compound of the invention or a prodrug of a compound of the invention to the individual in need.

Another aspect of the present invention is concerned with a method of inhibiting HCV NS5B polymerase, inhibiting HCV replication, or treating HCV infection with a compound of the present invention in combination with one or more agents useful for treating HCV infection. Such agents active against HCV include, but are not limited to, ribavirin, levovirin, viramidine, thymosin alpha-1, interferon-α, pegylated interferon-α (peginterferon-α), a combination of interferon-α and ribavirin, a combination of peginterferon-α and ribavirin, a combination of interferon-α and levovirin, and a combination of peginterferon-α and levovirin. Interferon-α includes, but is not limited to, recombinant interferon-α2a (such as Roferon interferon available from Hoffmann-

LaRoche, Nutley, N.J.), pegylated interferon-α2a (Pegasys™), interferon-α2b (such as Intron-A interferon available from Schering Corp., Kenilworth, N.J.), pegylated interferon-α2b (PegIntron™), a recombinant consensus interferon (such as interferon alphacon-1), and a purified interferon-α product. Amgen's recombinant consensus interferon has the brand name Infergen®. Levovirin is the L-enantiomer of ribavirin which has shown immunomodulatory activity similar to ribavirin. Viramidine is an amidino analog of ribavirin disclosed in WO 01/60379 (assigned to ICN Pharmaceuticals). In accordance with this method of the present invention, the individual components of the combination can be administered separately at different times during the course of therapy or concurrently in divided or single combination forms. The instant invention is therefore to be understood as embracing all such regimes of simultaneous or alternating treatment, and the term "administering" is to be interpreted accordingly. It will be understood that the scope of combinations of the compounds of this invention with other agents useful for treating HCV infection includes in principle any combination with any pharmaceutical composition for treating HCV infection. When a compound of the present invention or a pharmaceutically acceptable salt thereof is used in combination with a second therapeutic agent active against HCV, the dose of each compound may be either the same as or different from the dose when the compound is used alone.

For the treatment of HCV infection, the compounds of the present invention may also be administered in combination with an agent that is an inhibitor of HCV NS3 serine protease, such as LY570310 (VX-950). HCV NS3 serine protease is an essential viral enzyme and has been described to be an excellent target for inhibition of HCV replication. Both substrate and non-substrate based inhibitors of HCV NS3 protease inhibitors are disclosed in WO 98/17679, WO 98/22496, WO 98/46630, WO 99/07733, WO 99/07734, WO 99/38888, WO 99/50230, WO 99/64442, WO 00/09543, WO 00/59929, WO 01/74768, WO 01/81325, and GB-2337262. HCV NS3 protease as a target for the development of inhibitors of HCV replication and for the treatment of HCV infection is discussed in B. W. Dymock, "Emerging therapies for hepatitis C virus infection," *Emerging Drugs,* 6: 13-42 (2001).

Ribavirin, levovirin, and viramidine may exert their anti-HCV effects by modulating intracellular pools of guanine nucleotides via inhibition of the intracellular enzyme inosine monophosphate dehydrogenase (IMPDH). IMPDH is the rate-limiting enzyme on the biosynthetic route in de novo guanine nucleotide biosynthesis. Ribavirin is readily phosphorylated intracellularly and the monophosphate derivative is an inhibitor of IMPDH. Thus, inhibition of IMPDH represents another useful target for the discovery of inhibitors of HCV replication. Therefore, the compounds of the present invention may also be administered in combination with an inhibitor of IMPDH, such as VX-497, which is disclosed in WO 97/41211 and WO 01/00622, (assigned to Vertex); another IMPDH inhibitor, such as that disclosed in WO 00/25780 (assigned to Bristol-Myers Squibb); or mycophenolate mofetil [see A. C. Allison and E. M. Eugui, *Agents Action,* 44 (Suppl.): 165 (1993)].

For the treatment of HCV infection, the compounds of the present invention may also be administered in combination with the antiviral agent amantadine (1-aminoadamantane) [for a comprehensive description of this agent, see J. Kirschbaum, *Anal. Profiles Drug Subs.* 12: 1-36 (1983)].

The compounds of the present invention may also be combined for the treatment of HCV infection with antiviral 2'-C-branched ribonucleosides disclosed in R. E. Harry-O'kuru, et al., *J. Org. Chem.,* 62: 1754-1759 (1997); M. S. Wolfe, et al., *Tetrahedron Lett.,* 36: 7611-7614 (1995); and U.S. Pat. No. 3,480,613 (Nov. 25, 1969), the contents of which are incorporated by reference in their entirety. Such 2'-C-branched ribonucleosides include, but are not limited to, 2'-C-methyl-cytidine, 2'-C-methyl-adenosine, 2'-C-methyl-guanosine, and 9-(2-C-methyl-β-D-ribofuranosyl)-2,6-diaminopurine.

By "pharmaceutically acceptable" is meant that the carrier, diluent, or excipient must be compatible with the other ingredients of the formulation and not deleterious to the recipient thereof.

Also included within the present invention are pharmaceutical compositions comprising the novel nucleoside compounds and derivatives thereof of the present invention in association with a pharmaceutically acceptable carrier. Another example of the invention is a pharmaceutical composition made by combining any of the compounds described above and a pharmaceutically acceptable carrier. Another illustration of the invention is a process for making a pharmaceutical composition comprising combining any of the compounds described above and a pharmaceutically acceptable carrier.

Also included within the present invention are pharmaceutical compositions useful for inhibiting RNA-dependent RNA viral polymerase in particular HCV NS5B polymerase comprising an effective amount of a compound of this invention and a pharmaceutically acceptable carrier. Pharmaceutical compositions useful for treating RNA-dependent RNA viral infection in particular HCV infection are also encompassed by the present invention as well as a method of inhibiting RNA-dependent RNA viral polymerase in particular HCV NS5B polymerase and a method of treating RNA-dependent viral replication and in particular HCV replication. Additionally, the present invention is directed to a pharmaceutical composition comprising a therapeutically effective amount of a compound of the present invention in combination with a therapeutically effective amount of another agent active against RNA-dependent RNA virus and in particular against HCV. Agents active against HCV include, but are not limited to, ribavirin, levovirin, viramidine, thymosin alpha-1, an inhibitor of HCV NS3 serine protease, interferon-α, pegylated interferon-α (peginterferon-α), a combination of interferon-α and ribavirin, a combination of peginterferon-α and ribavirin, a combination of interferon-α and levovirin, and a combination of peginterferon-α and levovirin. Interferon-α includes, but is not limited to, recombinant interferon-α2a (such as Roferon interferon available from Hoffmann-LaRoche, Nutley, N.J.), interferon-α2b (such as Intron-A interferon available from Schering Corp., Kenilworth, N.J.), a consensus interferon, and a purified interferon-α product. For a discussion of ribavirin and its activity against HCV, see J. O. Saunders and S. A. Raybuck, "Inosine Monophosphate Dehydrogenase: Consideration of Structure, Kinetics, and Therapeutic Potential," *Ann. Rep. Med. Chem.,* 35: 201-210 (2000).

Another aspect of the present invention provides for the use of nucleoside compounds and derivatives thereof and their pharmaceutical compositions for the manufacture of a medicament for the inhibition of RNA-dependent RNA viral replication, in particular HCV replication, and/or the treatment of RNA-dependent RNA viral infection, in particular HCV infection. Yet a further aspect of the present invention provides for nucleoside compounds and derivatives thereof and their pharmaceutical compositions for use as a medicament for the inhibition of RNA-dependent RNA viral replication, in particular HCV replication, and/or for the treatment of RNA-dependent RNA viral infection, in particular HCV infection.

The pharmaceutical compositions of the present invention comprise a compound of structural formula I, IV, or XII as an active ingredient or a pharmaceutically acceptable salt thereof, and may also contain a pharmaceutically acceptable carrier and optionally other therapeutic ingredients.

The compositions include compositions suitable for oral, rectal, topical, parenteral (including subcutaneous, intramuscular, and intravenous), ocular (ophthalmic), pulmonary (nasal or buccal inhalation), or nasal administration, although the most suitable route in any given case will depend on the nature and severity of the conditions being treated and on the nature of the active ingredient. They may be conveniently presented in unit dosage form and prepared by any of the methods well-known in the art of pharmacy.

In practical use, the compounds of structural formulae I, IV, and XII can be combined as the active ingredient in intimate admixture with a pharmaceutical carrier according to conventional pharmaceutical compounding techniques. The carrier may take a wide variety of forms depending on the form of preparation desired for administration, e.g., oral or parenteral (including intravenous). In preparing the compositions for oral dosage form, any of the usual pharmaceutical media may be employed, such as, for example, water, glycols, oils, alcohols, flavoring agents, preservatives, coloring agents and the like in the case of oral liquid preparations, such as, for example, suspensions, elixirs and solutions; or carriers such as starches, sugars, microcrystalline cellulose, diluents, granulating agents, lubricants, binders, disintegrating agents and the like in the case of oral solid preparations such as, for example, powders, hard and soft capsules and tablets, with the solid oral preparations being preferred over the liquid preparations.

Because of their ease of administration, tablets and capsules represent the most advantageous oral dosage unit form in which case solid pharmaceutical carriers are obviously employed. If desired, tablets may be coated by standard aqueous or nonaqueous techniques. Such compositions and preparations should contain at least 0.1 percent of active compound. The percentage of active compound in these compositions may, of course, be varied and may conveniently be between about 2 percent to about 60 percent of the weight of the unit. The amount of active compound in such therapeutically useful compositions is such that an effective dosage will be obtained. The active compounds can also be administered intranasally as, for example, liquid drops or spray.

The tablets, pills, capsules, and the like may also contain a binder such as gum tragacanth, acacia, corn starch or gelatin; excipients such as dicalcium phosphate; a disintegrating agent such as corn starch, potato starch, alginic acid; a lubricant such as magnesium stearate; and a sweetening agent such as sucrose, lactose or saccharin. When a dosage unit form is a capsule, it may contain, in addition to materials of the above type, a liquid carrier such as a fatty oil.

Various other materials may be present as coatings or to modify the physical form of the dosage unit. For instance, tablets may be coated with shellac, sugar or both. A syrup or elixir may contain, in addition to the active ingredient, sucrose as a sweetening agent, methyl and propylparabens as preservatives, a dye and a flavoring such as cherry or orange flavor.

Compounds of structural formulae I, IV, and XII may also be administered parenterally. Solutions or suspensions of these active compounds can be prepared in water suitably mixed with a surfactant such as hydroxy-propylcellulose. Dispersions can also be prepared in glycerol, liquid polyethylene glycols and mixtures thereof in oils. Under ordinary conditions of storage and use, these preparations contain a preservative to prevent the growth of microorganisms.

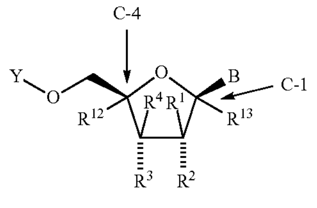
The pharmaceutical forms suitable for injectable use include sterile aqueous solutions or dispersions and sterile powders for the extemporaneous preparation of sterile injectable solutions or dispersions. In all cases, the form must be sterile and must be fluid to the extent that easy syringability exists. It must be stable under the conditions of manufacture and storage and must be preserved against the contaminating action of microorganisms such as bacteria and fungi. The carrier can be a solvent or dispersion medium containing, for example, water, ethanol, polyol (e.g. glycerol, propylene glycol and liquid polyethylene glycol), suitable mixtures thereof, and vegetable oils.

Any suitable route of administration may be employed for providing a mammal, especially a human with an effective dosage of a compound of the present invention. For example, oral, rectal, topical, parenteral, ocular, pulmonary, nasal, and the like may be employed. Dosage forms include tablets, troches, dispersions, suspensions, solutions, capsules, creams, ointments, aerosols, and the like. Preferably compounds of structural formulae I, IV, and XII are administered orally.

For oral administration to humans, the dosage range is 0.01 to 1000 mg/kg body weight in divided doses. In one embodiment the dosage range is 0.1 to 100 mg/kg body weight in divided doses. In another embodiment the dosage range is 0.5 to 20 mg/kg body weight in divided doses. For oral administration, the compositions are preferably provided in the form of tablets or capsules containing 1.0 to 1000 milligrams of the active ingredient, particularly, 1, 5, 10, 15, 20, 25, 50, 75, 100, 150, 200, 250, 300, 400, 500, 600, 750, 800, 900, and 1000 milligrams of the active ingredient for the symptomatic adjustment of the dosage to the patient to be treated.

The effective dosage of active ingredient employed may vary depending on the particular compound employed, the mode of administration, the condition being treated and the severity of the condition being treated. Such dosage may be ascertained readily by a person skilled in the art. This dosage regimen may be adjusted to provide the optimal therapeutic response.

The compounds of the present invention contain one or more asymmetric centers and can thus occur as racemates and racemic mixtures, single enantiomers, diastereomeric mixtures and individual diastereomers. The present invention is meant to comprehend nucleoside derivatives having the β-D stereochemical configuration for the five-membered furanose ring as depicted in the structural formula below, that is, nucleoside compounds in which the substituents at C-1 and C-4 of the five-membered furanose ring have the (3-stereochemical configuration ("up" orientation as denoted by a bold line).

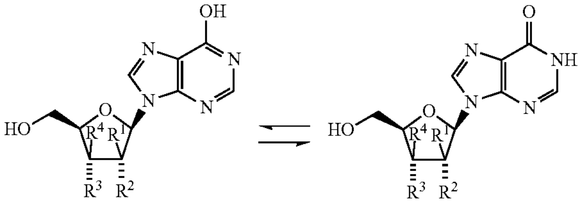


β-D-

The stereochemistry of the substituents at the C-2 and C-3 positions of the furanose ring of the compounds of the present invention is denoted either by a dashed line which signifies that the substituent, for example R^2 in structural formula VI, has the α (substituent "down") configuration or a squiggly line which signifies that the substituent, for example R^3 in structural formula VI, can have either the α (substituent "down") or β (substituent "up") configuration.

Some of the compounds described herein contain olefinic double bonds, and unless specified otherwise, are meant to include both E and Z geometric isomers.

Some of the compounds described herein may exist as tautomers such as keto-enol tautomers. The individual tautomers as well as mixtures thereof are encompassed with compounds of structural formulae I, IV, and XII. An example of keto-enol tautomers which are intended to be encompassed within the compounds of the present invention is illustrated below:

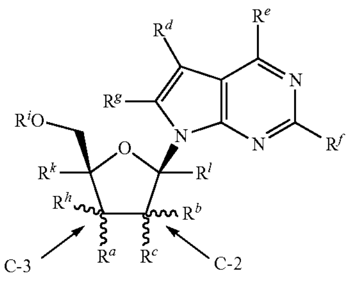


Compounds of structural formulae I, IV, and XII may be separated into their individual diastereoisomers by, for example, fractional crystallization from a suitable solvent, for example methanol or ethyl acetate or a mixture thereof, or via chiral chromatography using an optically active stationary phase.

Alternatively, any stereoisomer of a compound of the structural formulae I, IV, and XII may be obtained by stereospecific synthesis using optically pure starting materials or reagents of known configuration.

The stereochemistry of the substituents at the C-2 and C-3 positions of the furanose ring of the novel compounds of the present invention of structural formula XII is denoted by squiggly lines which signifies that substituents R^a, R^b, R^c and R^h can have either the I (substituent "down") or θ (substituent "up") configuration independently of one another.

(XII)



The compounds of the present invention may be administered in the form of a pharmaceutically acceptable salt. The term "pharmaceutically acceptable salt" refers to salts prepared from pharmaceutically acceptable non-toxic bases or acids including inorganic or organic bases and inorganic or organic acids. Salts of basic compounds encompassed within the term "pharmaceutically acceptable salt" refer to non-toxic salts of the compounds of this invention which are generally prepared by reacting the free base with a suitable organic or inorganic acid. Representative salts of basic compounds of the present invention include, but are not limited to, the following: acetate, benzenesulfonate, benzoate, bicarbonate, bisulfate, bitartrate, borate, bromide, camsylate, carbonate, chloride, clavulanate, citrate, dihydrochloride, edetate, edisylate, estolate, esylate, fumarate, gluceptate, gluconate, glutamate, glycollylarsanilate, hexylresorcinate, hydrabamine, hydrobromide, hydrochloride, hydroxynaphthoate, iodide, isothionate, lactate, lactobionate, laurate, malate, maleate, mandelate, mesylate, methylbromide, methylnitrate, methylsulfate, mucate, napsylate, nitrate, N-methylglucamine ammonium salt, oleate, oxalate, pamoate (embonate), palmitate, pantothenate, phosphate/diphosphate, polygalacturonate, salicylate, stearate, sulfate, subacetate, succinate, tannate, tartrate, teoclate, tosylate, triethiodide and valerate. Furthermore, where the compounds of the invention carry an acidic moiety, suitable pharmaceutically acceptable salts thereof include, but are not limited to, salts derived from inorganic bases including aluminum, ammonium, calcium, copper, ferric, ferrous, lithium, magnesium, manganic, mangamous, potassium, sodium, zinc, and the like. Particularly preferred are the ammonium, calcium, magnesium, potassium, and sodium salts. Salts derived from pharmaceutically acceptable organic non-toxic bases include salts of primary, secondary, and tertiary amines, cyclic amines, and basic ion-exchange resins, such as arginine, betaine, caffeine, choline, N,N-dibenzylethylenediamine, diethylamine, 2-diethylaminoethanol, 2-dimethylaminoethanol, ethanolamine, ethylenediamine, N-ethylmorpholine, N-ethylpiperidine, glucamine, glucosamine, histidine, hydrabamine, isopropylamine, lysine, methylglucamine, morpholine, piperazine, piperidine, polyamine resins, procaine, purines, theobromine, triethylamine, trimethylamine, tripropylamine, tromethamine, and the like.

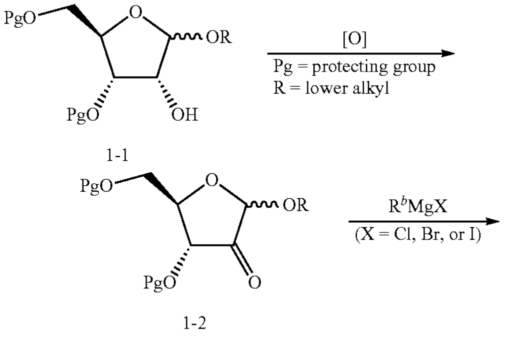
Also, in the case of a carboxylic acid (—COOH) or alcohol group being present in the compounds of the present invention, pharmaceutically acceptable esters of carboxylic acid derivatives, such as methyl, ethyl, or pivaloyloxymethyl, or acyl derivatives of alcohols, such as acetate or maleate, can be employed. Included are those esters and acyl groups known in the art for modifying the solubility or hydrolysis characteristics for use as sustained-release or prodrug formulations.

Preparation of the Nucleoside Compounds and Derivatives of the Invention

The nucleoside compounds and derivatives thereof of the present invention can be prepared following synthetic methodologies well-established in the practice of nucleoside and nucleotide chemistry. Reference is made to the following text for a description of synthetic methods used in the preparation of the compounds of the present invention: "Chemistry of Nucleosides and Nucleotides," L. B. Townsend, ed., Vols. 1-3, Plenum Press, 1988, which is incorporated by reference herein in its entirety.

A representative general method for the preparation of compounds of the present invention is outlined in Scheme 1 below. This scheme illustrates the synthesis of compounds of the present invention of structural formula 1-7 wherein the furanose ring has the O-D-ribo configuration. The starting material is a 3,5-bis-O-protected alkyl furanoside, such as methyl furanoside, of structural formula 1-1. The C-2 hydroxyl group is then oxidized with a suitable oxidizing agent, such as a chromium trioxide or chromate reagent or Dess-Martin periodinane, or by Swern oxidation, to afford a C-2 ketone of structural formula 1-2. Addition of a Grignard reagent, such as an alkyl, alkenyl, or alkynyl magnesium halide (for example, MeMgBr, EtMgBr, vinylMgBr, allylMgBr, and ethynylMgBr) or an alkyl, alkenyl, or alkynyl lithium, such as MeLi, across the carbonyl double bond of 1-2 in a suitable organic solvent, such as tetrahydrofuran, diethyl ether, and the like, affords the C-2 tertiary alcohol of structural formula 1-3. A good leaving group (such as Cl, Br, and I) is next introduced at the C-1 (anomeric) position of the furanose sugar derivative by treatment of the furanoside of formula 1-3 with a hydrogen halide in a suitable organic solvent, such as hydrogen bromide in acetic acid, to afford the intermediate furanosyl halide 1-4. A C-1 sulfonate, such methanesulfonate ($MeSO_2O$—), trifluoromethanesulfonate (CF_3SO_2O—), or p-toluenesulfonate (—OTs), may also serve as a useful leaving group in the subsequent reaction to generate the glycosidic (nucleosidic) linkage. The nucleosidic linkage is constructed by treatment of the intermediate of structural formula 1-4 with the metal salt (such as lithium, sodium, or potassium) of an appropriately substituted 1H-pyrrolo[2,3-d]pyrimidine 1-5, such as an appropriately substituted 4-halo-1H-pyrrolo[2,3-d]pyrimidine, which can be generated in situ by treatment with an alkali hydride (such as sodium hydride), an alkali hydroxide (such as potassium hydroxide), an alkali carbonate (such as potassium carbonate), or an alkali hexamethyldisilazide (such as NaHMDS) in a suitable anhydrous organic solvent, such as acetonitrile, tetrahydrofuran, 1-methyl-2-pyrrolidinone, or N,N-dimethylformamide (DMF). The displacement reaction can be catalyzed by using a phase-transfer catalyst, such as TDA-1 or triethylbenzylammonium chloride, in a two-phase system (solid-liquid or liquid-liquid). The optional protecting groups in the protected nucleoside of structural formula 1-6 are then cleaved following established deprotection methodologies, such as those described in T. W. Greene and P. G. M. Wuts, "Protective Groups in Organic Synthesis," 3^{rd} ed., John Wiley & Sons, 1999. Optional introduction of an amino group at the 4-position of the pyrrolo[2,3-d]pyrimidine nucleus is effected by treatment of the 4-halo intermediate 1-6 with the appropriate amine, such as alcoholic ammonia or liquid ammonia, to generate a primary amine at the C-4 position (—NH_2), an alkylamine to generate a secondary amine (—NHR), or a dialkylamine to generate a tertiary amine (—NRR'). A 7H-pyrrolo[2,3-d]pyrimidin-4(3H) one compound may be derived by hydrolysis of 1-6 with aqueous base, such as aqueous sodium hydroxide. Alcoholysis (such as methanolysis) of 1-6 affords a C_{1-4} alkoxide (—OR), whereas treatment with an alkyl mercaptide affords a C-4 alkylthio (—SR) derivative. Subsequent chemical manipulations well-known to practitioners of ordinary skill in the art of organic/medicinal chemistry may be required to attain the desired compounds of the present invention.

Scheme 1



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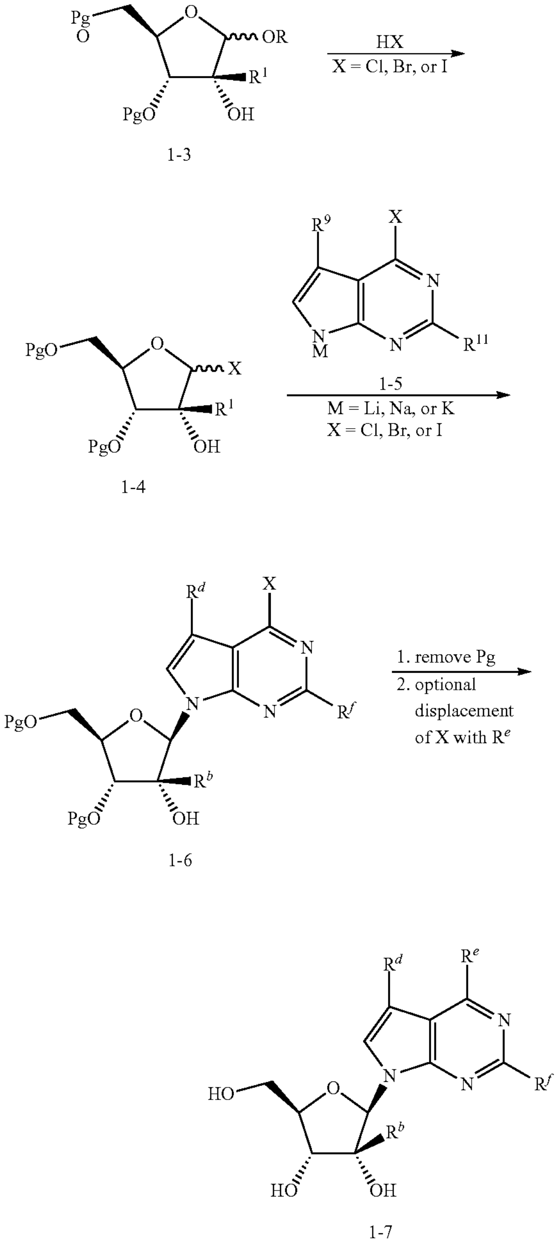
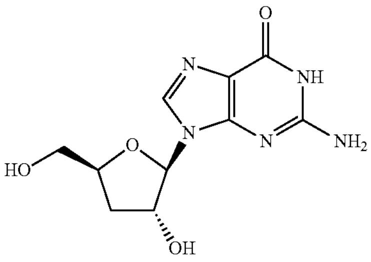


The examples below provide citations to literature publications, which contain details for the preparation of final compounds or intermediates employed in the preparation of final compounds of the present invention. The nucleoside compounds of the present invention were prepared according to procedures detailed in the following examples. The examples are not intended to be limitations on the scope of the instant invention in any way, and they should not be so construed. Those skilled in the art of nucleoside and nucleotide synthesis will readily appreciate that known variations of the conditions and processes of the following preparative procedures can be used to prepare these and other compounds of the present invention. All temperatures are degrees Celsius unless otherwise noted.

EXAMPLE 1

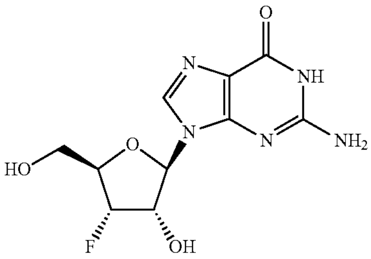
3'-Deoxyguanosine



This compound was prepared following the procedures described in *Nucleosides Nucleotides,* 13: 1049 (1994).

EXAMPLE 2

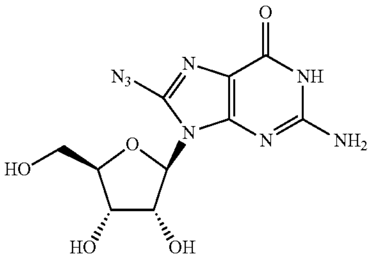
3'-Deoxy-3'-fluoroguanosine



This compound was prepared following the procedures described in *J. Med. Chem.* 34: 2195 (1991).

EXAMPLE 3

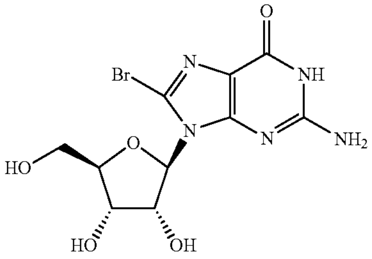
8-Azidoguanosine



This compound was prepared following the procedures described in *Chem. Pharm. Bull.* 16: 1616 (1968).

EXAMPLE 4

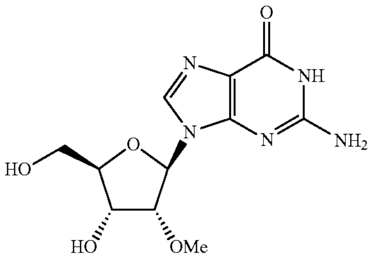
8-Bromoguanosine



This compound was obtained from commercial sources.

EXAMPLE 5

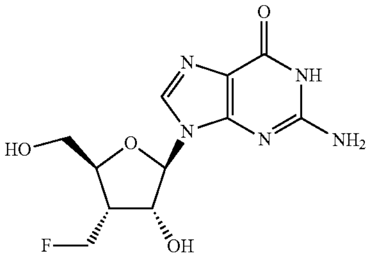
2'-O-Methylguanosine



This compound was obtained from commercial sources.

EXAMPLE 6

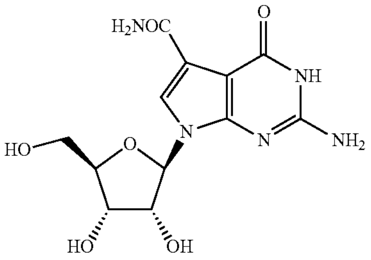
3'-Deoxy-3'-(fluoromethyl)guanosine



To a solution of 1,2-O-diacetyl-5-O-(p-toluoyl)-3-deoxy-3-(fluoromethyl)-D-ribofuranose (257 mg, 0.7 mmol) [prepared by a similar method as that described for the corresponding 5-O-benzyl derivative in *J. Med. Chem.* 36: 353 (1993)] and N^2-acetyl-O^6-(diphenylcarbamoyl)guanine (554 mg, 1.43 mmol) in anhydrous acetonitrile (6.3 mL) was added bis(trimethylsilyl)acetamide (BSA) (1.03 g, 5 mmol). The reaction mixture was stirred at reflux for 30 minutes, and the bath was removed. The reaction mixture was cooled in an ice bath and TMS-triflate (288 mg, 1.3 mmol) was added with stirring. After addition was complete, the reaction was heated at reflux for 2 hr., the reaction mixture was poured onto ice and extracted with chloroform (5×10 mL). The combined organic layers were washed with aqueous saturated sodium bicarbonate, brine and dried over anhydrous Na_2SO_4. The solvent was removed under reduced pressure and the residue chromatographed over silica gel using 5% acetone/CH_2Cl_2 as the eluant to furnish the fully protected corresponding nucleoside derivative. This was dissolved in 1,4-dioxane (1.5 mL) to which was added 40% $MeNH_2/H_2O$ (1.3 g, 17 mmol). The reaction mixture was stirred for 1 day, evaporated and the residue crystallized with ether/MeOH to provide the title compound (58 mg). ^1H NMR (DMSO-d_6): δ 2.76-2.67 (m, 1H); 3.55-3.50 (m, 1H), 2.76-2.67 (m, 1H); 3.71-3.66 (m, 1H), 4.08-4.04 (m, 1H), 4.77-4.50 (m, 3H), 5.06 (t, 1H, J=5.3 Hz), 5.69 (d, 1H, J=3.4 Hz), 5.86 (d, 1H, J=5.1 Hz), 6.45 (bs, 2H), 7.97 (s, 1H), 10.59 (s, 1H). ^{19}F NMR (DMSO-d_6): δ −221.46 (m, F).

EXAMPLE 7

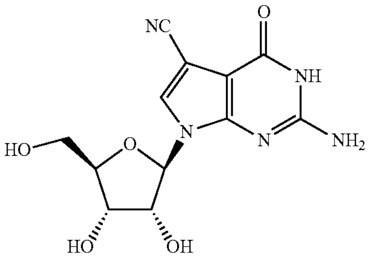
2-Amino-3,4-dihydro-4-oxo-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide



This compound was prepared following the procedures described in *Tetrahedron. Lett.* 25: 4793 (1983).

EXAMPLE 8

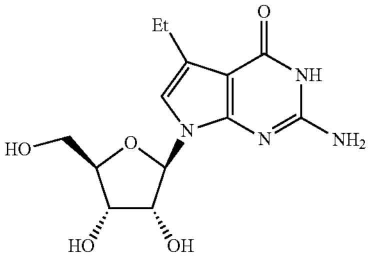
2-Amino-3,4-dihydro-4-oxo-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile



This compound was prepared following the procedures described in *J. Am. Chem. Soc.* 98: 7870 (1976).

EXAMPLE 9

2-Amino-5-ethyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



Step A: 2-Amino-7-(5-O-tert-butyldimethylsilyl-2,3-O-isopropylidene-β-D-ribofuranosyl)-4-chloro-5-ethyl-7H-pyrrolo[2,3-d]pyrimidine

To a stirred suspension of 2-amino-4-chloro-5-ethyl-1H-pyrrolo[2,3-d]pyrimidine [described in EP 866070 (1998)] (1.57 g, 8 mmol) in dry MeCN (48 mL) was added NaH (60% in mineral oil; 0.32 g, 8 mmol), and the mixture was stirred at room temperature for 1 h. A solution of 5-O-tert-butyldimethylsilyl-2,3-O-isopropylidene-α-D-ribofuranosyl chloride [generated in situ from the corresponding lactol (1.95 g, 6.4 mmol) according to Wilcox et al., *Tetrahedron Lett.,* 27: 1011 (1986)] in dry THF (9.6 mL) was added at room temperature, and the mixture was stirred overnight, then evaporated to dryness. The residue was suspended in water (100 mL) and extracted with EtOAc (200+150 mL). The combined extracts were washed with brine, dried (Na_2SO_4) and evaporated. The residue was purified on a silica gel column using a solvent system of hexanes/EtOAc:7/1. Appropriate fractions were collected and evaporated to dryness to give the title compound (1.4 g) as a colorless foam.

Step B: 2-Amino-4-chloro-5-ethyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

A mixture of the compound from Step A (1.19 g, 2.5 mmol) in MeOH (100 mL) and water (50 mL) was stirred with DOWEX H^+ (to adjust pH of the mixture to 5) at room temperature for 2.5 h. The mixture was filtered and the resin thoroughly washed with MeOH. The combined filtrate and washings were evaporated and the residue coevaporated several times with water to yield the title compound (0.53 g) as a white solid.

Step C: 2-Amino-5-ethyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one

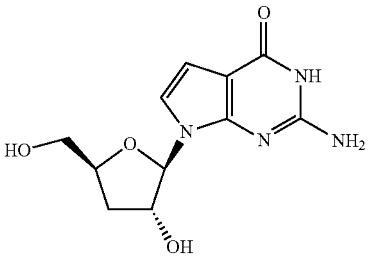
A mixture of the compound from Step B (104 mg, 0.32 mmol) in 2N aqueous NaOH (10 mL) was stirred at reflux temperature for 15 min. The solution was cooled in ice bath, neutralized with 2 N aqueous HCl, and evaporated to dryness. The residue was suspended in MeOH, mixed with silica gel, and evaporated. The solid residue was placed onto a silica gel column (packed in a solvent mixture of CH_2Cl_2/MeOH: 10/1) which was eluted with a solvent system of CH_2Cl_2/MeOH:

10/1 and 5/1. The fractions containing the product were collected and evaporated to dryness to yield the title compound (48 mg) as a white solid.

^{1}H NMR (CD_3OD): δ 1.22 (t, 3H), 2.69 (q, 2H), 3.69, 3.80 (2m, 2H), 4.00 (m, 1H), 4.22 (m, 1H), 4.45 (t, 1H), 5.86 (d, 1H, J=6.0 Hz), 6.60 (d, 1H, J=1.2 Hz).

EXAMPLE 10

2-Amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



Step A: 2-Amino-7-(2,3-anhydro-β-D-ribofuranosyl)-4-methoxy-7H-pyrrolo[2,3-d]pyrimidine

To a mixture of 2-amino-7-(β-D-ribofuranosyl)-4-chloro-7H-pyrrolo[2,3-c]pyrimidine (1.8 g, 6.0 mmol) in acetonitrile (80 mL) were added a solution of H_2O/CH_3CN (1:9, 1.08 mL) and then α-acetoxyisobutyryl bromide (3.5 mL, 24 mmol). After 2 h stirring at room temperature, saturated aqueous $NaHCO_3$ (170 mL) was added and the mixture was extracted with EtOAc (300+200 mL). The combined organic phase was washed with brine (100 mL), dried (Na_2SO_4) and evaporated to a pale yellow foamy residue. This was suspended in anhydrous MeOH (80 mL) and stirred overnight with 25 mL of DOWEX Off resin (previously washed with anhydrous MeOH). The resin was filtered, washed thoroughly with MeOH and the combined filtrate evaporated to give a pale yellow foam (1.92 g).

Step B: 2-Amino-7-(3-deoxy-β-D-ribofuranosyl)-4-methoxy-7H-pyrrolo[2,3-d]pyrimidine

A solution of $LiEt_3BH/THF$ (1M, 75 mL, 75 mmol) was added dropwise to a cold (ice bath) deoxygenated (Ar, 15 min) solution of the compound from Step A (1.92 g) under Ar. Stirring at 0° C. was continued for 4 h. At this point the reaction mixture was acidified with 5% aqueous acetic acid (110 mL), then purged with Ar for 1 h and finally evaporated to a solid residue. Purification on a silica gel column using $MeOH/CH_2Cl_2$ as eluent yielded target compound as a colourless foam (1.01 g).

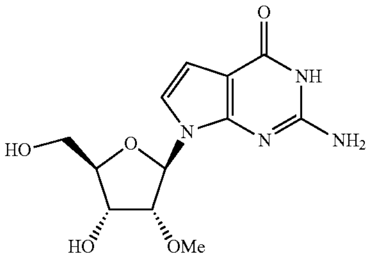
Step C: 2-Amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-4(3H)-one

A mixture of compound from Step B (0.4 g, 1.4 mmol) in 2N aqueous NaOH (40 mL) was stirred at reflux temperature for 3 h. The solution was cooled in ice bath, neutralized with 2 N aqueous HCl and evaporated to dryness. The residue was suspended in MeOH, mixed with silica and evaporated. The residue was placed onto a silica gel column which was eluted with CH_2Cl_2/MeOH: 10/1 and 5/1 to give the title compound as white solid (0.3 g).

^{1}H NMR (DMSO-d_6): δ 1.85, 2.12 (2m, 2H), 3.55, 3.46 (2dd, 2H), 4.18 (m, 1H); 4.29 (m, 1H), 4.85 (7, 1H), 5.42 (d, 1H) 5.82 (d, 1H, J=2.4 Hz), 6.19 (s, 2H), 6.23 (d, 1H, J=3.6 Hz), 6.87 (d, 1H), 10.31 (s, 1H).

EXAMPLE 11

2-Amino-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



Step A: 2-Amino-4-chloro-7-(5-t-butyldimethylsilyl-2,3-O-isopropylidene-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

HMPT (10.65 ml, 55 mmol) was added portionwise over 30 min. to a solution of 5-O-tert-butyldimethylsilyl-2,3-O-isopropylidene-D-ribofuranose (13.3 g, 44 mmol), dry THF (135 mL), CCl_4 (5.62 mL, 58 mmol) under N_2 at −76° C. After 30 min., the temp. was raised to −20° C. In a separate flask, a suspension of 2-amino-4-chloro-1H-pyrrolo-[2,3-d]-pyrimidine (15 g, 89 mmol) in CH_3CN (900 mL) was treated at 15° C. with 60% NaH (3.60 g., 90 mmol.). The reaction was stirred 30 min. whereupon the previous reaction mixture was cannulated with vigorous stirring. The reaction was stirred 16 hrs. and then concentrated in vacuo. The resulting semisolid was added to ice/water/EtOAc and extracted with EtOAc (3×200 mL), dried $NaSO_4$, filtered and evaporated. The resulting oil was chromatographed on silica gel (EtOAc/Hexane 1/1) to afford the product as an oil (9.0 g).

Step B: 2-Amino-4-chloro-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

A solution of the compound from Step A (5.76 g, 13 mmol) in $MeOH/H_2O$ (1200 mL/600 mL) and Dowex WX8-400 (4.8 g) was stirred 16 hrs. at room temperature. The resin was filtered off and the filtrate evaporated to afford the title compound as a white solid; yield 3.47 g.

^{1}H NMR (DMSO-d_6): δ 3.56 (m, 2H), 3.86 (m, 1H), 4.07 (m, 1H), 4.32 (m, 1H), 4.99 (t, 1H), 5.10 (d, 1H), 5.30 (d, 1H), 6.00 (d, 1H), 6.38 (d, 1H), 6.71 (s br, 2H), 7.39 (d, 1H).

Step C: 2-Amino-4-chloro-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

A solution of the compound from Step B (1.0 g, 3.3 mmol) in dry DMF (100 mL) at 15° C. was treated with 60% NaH (0.14 g, 3.5 mmol). After 30 min., iodomethane (47 g, 3.3 mmol) was added portionwise to the stirred solution. The reaction was stirred at room temperature for 16 hrs. and then evaporated at a temperature below 40° C. The resulting solid was chromatographed on silica gel to afford the product as a white solid; yield 0.81 g. ^{1}H NMR (DMSO-d_6): δ 3.25 (s,

3H), 3.54 (m, 2H), 3.87 (m, 1H), 4.07 (m, 1H), 4.22 (m, 1H), 5.01 (m, 1H), 5.16 (d, 1H), 6.07 (d, 1H), 6.37 (d, 1H), 6.70 (s br, 2H), 7.40 (s, 1H). Mass spectrum: m/z 316 (M+1)$^+$.

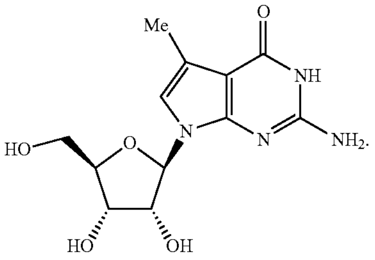
Step D: 2-Amino-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one

A solution of the compound from Step C (80 mg, 0.25 mmol) in NaOH/H_2O (1.6 g/20 ml) was heated at reflux for 7 hrs., whereupon the solution was adjusted with dilute HCl to a pH of 7 and then evaporated. Chromatography of the resulting solid on silica gel with EtOAc/MeOH 8/2 afforded the product as a white solid; yield 64 mg.

^{1}H NMR (DMSO-d_6): δ 3.25 (s, 3H), 3.52 (m, 2H) 3.81 (m, 1H), 4.00 (m, 1H), 4.19 (m, 1H), 5.10 (s br, 2H), 5.95 (d, 1H), 6.27 (d, 1H), 6.33 (s br, 2H), 6.95 (d, 1H), 10.55 (s br, 1H).

EXAMPLE 12

2-Amino-5-methyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



This compound is described in *Biochemistry,* 33: 2703 (1994) and was synthesized by the following procedure:

Step A: 2-Amino-7-(5-O-tert-butyldimethylsilyl-2,3-O-isopropylidene-β-D-ribofuranosyl)-4-chloro-5-methyl-7H-pyrrolo[2,3-d]pyrimidine

To a stirred suspension of 2-amino-4-chloro-5-methyl-1H-pyrrolo[2,3-d]pyrimidine (*Liebigs Ann. Chem.* 1984, 4, 708) (0.91 g, 5 mmol) in dry MeCN (30 ml) was added NaH (60% in mineral oil; 0.2 g, 5 mmol) and the mixture was stirred at room temperature for 0.5 h. A solution of 5-O-tert-butyldimethylsilyl-2,3-O-isopropylidene-α-D-ribofuranosyl chloride [generated in situ from the corresponding lactol (1.22 g, 4 mmol) according to *Tetrahedron Lett.* 27: 1011 (1986)] in dry THF (6 mL) was added at room temperature, and the mixture was stirred overnight, then evaporated to dryness. The residue was suspended in water (100 mL) and extracted with EtOAc (2×100 mL). The combined organic extracts were washed with brine, dried (Na_2SO_4) and evaporated. The residue was purified on a silica gel column using a solvent system of hexanes/EtOAc:7/1 and 5/1. Appropriate fractions were collected and evaporated to dryness to give the title compound (0.7 g) as a colorless foam.

Step B: 2-Amino-4-chloro-5-methyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

A mixture of the intermediate from Step A (0.67 g, 1.4 mmol) in MeOH (70 ml) and water (35 ml) was stirred with DOWEX H$^+$ (to adjust pH of the mixture to 5) at room temperature for 4 h. The mixture was filtered and the resin thoroughly washed with MeOH. The combined filtrate and washings were evaporated and the residue coevaporated several times with water to yield the title compound (0.37 g) as a white solid.

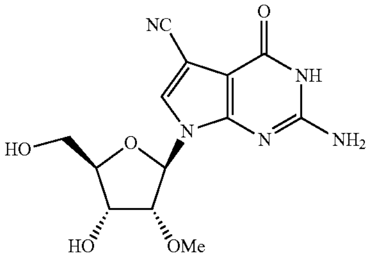
Step C: 2-Amino-5-methyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one

A mixture of intermediate from Step B (100 mg, 0.32 mmol) in 2N aqueous NaOH (20 mL) was stirred at reflux temperature for 1.5 h. The solution was cooled in ice bath, neutralized with 2 N aqueous HCl and evaporated to dryness. The residue was suspended in MeOH, mixed with silica gel and evaporated. The solid residue was placed onto a silica gel column (packed in a solvent mixture of CH_2Cl_2/MeOH: 10/1) which was eluted with a solvent system of CH_2Cl_2/MeOH: 10/1 and 5/1. The fractions containing the product were collected and evaporated to dryness to yield the title compound (90 mg) as a white solid.

^{1}H NMR (DMSO-d_6): δ 2.15 (d, 3H), 3.47, 3.50 (2m, 2H), 3.75 (m, 1H), 3.97 (m, 1H), 4.17 (m, 1H), 4.89 (t, 1H), 4.96 (d, 1H), 5.14 (d, 1H), 5.80 (d, 1H, J=6.4 Hz), 6.14 (s, 2H), 6.60 (q, 1H, J=1.2 Hz), 10.23 (s, 1H).

EXAMPLE 13

2-Amino-3,4-dihydro-4-oxo-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile



Step A: 2-Amino-4-chloro-7-β-D-ribofuranosyl-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile

This intermediate was prepared according to *J. Chem. Soc. Perkin Trans.* 1. 2375 (1989).

Step B: 2-Amino-4-chloro-7-[3,5-O-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile

To a solution of the compound from Step A (1.64 g, 5.00 mmol) in DMF (30 mL) was added imidazole (0.681 g, 10.0 mmol). The solution was cooled to 0° C. and 1,3-dichloro-1,1,3,3-tetraisopropyldisiloxane (1.58 g, 5.00 mmol) was added dropwise. The bath was removed and the solution stirred at room temperature for 30 minutes, evaporated in vacuo to an oil, taken up in ethyl acetate (150 mL) and washed with saturated aqueous sodium bicarbonate (50 mL) and with water (50 mL). The organic phase was dried over magnesium sulfate, filtered and evaporated in vacuo. The residue was purified on silica gel using ethyl acetate/hexane (1:2) as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (2.05 g) as a colorless foam.

^{1}H NMR (DMSO-d$_6$): δ 1.03 (m, 28H), 3.92 (m, 1H), 4.01 (m, 1H), 4.12 (m, 1H), 4.24 (m, 2H), 5.67 (m, 1H), 5.89 (s, 1H), 7.17 (bs, 2H), 8.04 (s, 1H).

Step C: 2-Amino-4-chloro-7-[2-O-methyl-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile

To a pre-cooled solution (0° C.) of the compound from Step B (1.70 g, 3.00 mmol) in DMF (30 mL) was added methyl iodide (426 mg, 3.00 mmol) and then NaH (60% in mineral oil) (120 mg, 3.00 mmol). The mixture was stirred at rt for 30 minutes and then poured into a stirred mixture of saturated aqueous ammonium chloride (100 mL) and ethyl acetate (100 mL). The organic phase was washed with water (100 mL), dried over magnesium sulfate, filtered and evaporated in vacuo. The resulting oily residue was co-evaporated three times from acetonitrile (10 mL), taken up in THF (50 mL) and tetrabutylammonium fluoride (1.1 mmol/g on silica) (4.45 g, 6.00 mmol) was added. The mixture was stirred for 30 minutes, filtered and the filtrate evaporated in vacuo. The crude product was purified on silica using methanol/dichloromethane (7:93) as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (359 mg) as a colorless solid.

^{1}H NMR (DMSO-d$_6$): δ 3.30 (s, 3H), 3.56 (m, 2H) 3.91 (m, 1H), 4.08 (m, 1H), 4.23 (m, 1H), 5.11 (m, 1H), 5.23 (m, 1H), 7.06 (m, 1H), 7.16 (bs, 2H), 8.38 (s, 1H).

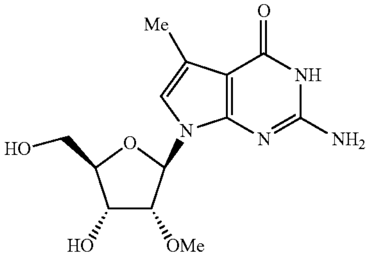
Step D: 2-Amino-3,4-dihydro-4-oxo-7-[2-O-methyl-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile

To a solution of the compound from Step D in DMF (5.0 mL) and dioxane (3.5 mL) was added syn-pyridinealdoxime (336 mg, 2.75 mmol) and then tetramethylguanidine (288 mg, 2.50 mmol). The resulting solution was stirred overnight at rt, evaporated in vacuo and co-evaporated three times from acetonitrile (20 mL). The oily residue was purified on silica gel using methanol/dichloromethane (7:93) as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (103 mg) as a colorless solid.

^{1}H NMR (DMSO-d$_6$): δ 3.30 (s, 3H), 3.57 (m, 2H), 3.86 (m, 1H), 4.00 (m, 1H), 4.21 (m, 1H), 5.07 (m, 1H), 5.17 (m, 1H), 5.94 (m, 1H), 6.56 (bs, 2H), 7.93 (s, 1H), 10.82 (bs, 1H).

EXAMPLE 14

2-Amino-5-methyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



Step A: 2-Amino-4-chloro-5-methyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]-pyrimidine

Into a solution of the compound from Example 12, Step B (188 mg, 0.6 mmol) in anhydrous DMF (6 mL) was added NaH (60% in mineral oil; 26 mg, 0.66 mmol). The mixture was stirred at room temperature for 0.5 h and then cooled. MeI (45 μL) was added at 0° C. and the reaction mixture allowed to warm to 15° C. in 5 h. Then the mixture was poured into ice-water (20 mL) and extracted with CH_2Cl_2 (100+50 mL). The combined organic extracts were washed with water (50 mL), brine (50 mL) and dried (Na_2SO_4). The evaporated residue was purified on a silica gel column with a solvent system of CH_2Cl_2/MeOH: 30/1. Appropriate fractions were pooled and evaporated to yield the title compound (50 mg) as a colorless glass.

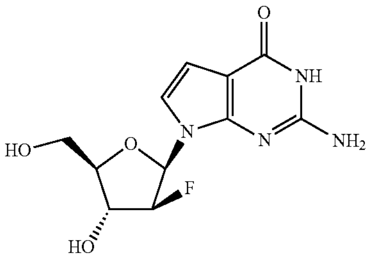
Step B: 2-Amino-7-(2-O-methyl-β-D-ribofuranosyl)-5-methyl-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one

A solution of the compound from Step A (50 mg, 0.15 mmol) in 0.5M NaOMe/MeOH (4 mL) was stirred at reflux temperature for 1.5 h. The mixture was cooled, mixed with silica gel and evaporated to dryness. The silica gel was loaded onto a silica gel column and eluted with a solvent system of CH_2Cl_2/MeOH: 30/1. The fractions containing the product were collected and evaporated to yield 2-amino-7-(2-O-methyl-β-D-ribofuranosyl)-4-methoxy-5-methyl-7H-pyrrolo[2,3-d]pyrimidine (40 mg). This was mixed with 2 N aqueous NaOH (4 mL) and stirred at reflux temperature for 10 h. The mixture was cooled in ice bath, neutralized with 2 N aqueous HCl and evaporated. The solid residue was suspended in MeOH, mixed with silica gel and evaporated. The silica gel was loaded onto a silica gel column and eluted with a solvent system of CH_2Cl_2/MeOH: 5/1. Appropriate fractions were pooled and evaporated to give the title compound (40 mg) as a white solid.

^{1}H NMR (DMSO-d$_6$): δ 2.18 (s, 3H), 3.26 (s, 3H), 3.45, 3.52 (2m, 2H), 3.82 (m, 1H), 3.97 (dd, 1H), 4.20 (m, 1H), 4.99 ((t, 1H), 5.10 (d, 1H), 5.94 (d, 1H, J=7.0 Hz), 6.19 (bs, 2H), 6.68 (s, 1H), 10.60 (br, 1H).

EXAMPLE 15

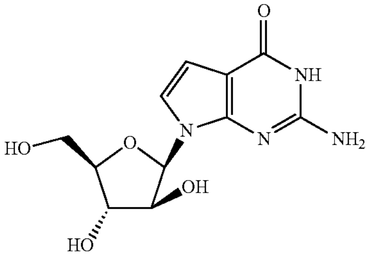
2-Amino-7-(2-deoxy-2-fluoro-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



This compound was prepared following the procedures described in *J. Med. Chem.* 38: 3957 (1995).

EXAMPLE 16

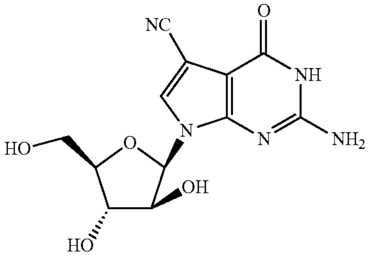
2-Amino-7-(β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



This compound was prepared following the procedures described in *J. Org. Chem.* 47: 226 (1982).

EXAMPLE 17

2-Amino-7-(β-D-arabinofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile



Step A: 2-Amino-7-(β-D-arabinofuranosyl)-4-chloro-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile

This intermediate was prepared according to *J. Chem. Soc. Perkin Trans.* 1, 2375 (1989).

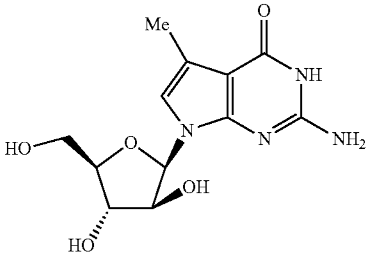
Step B: 2-Amino-7-(β-D-arabinofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile

To a solution of the compound from Step A (163 mg, 0.50 mmol) in DMF (5.0 mL) and dioxane (3.5 mL) was added syn-pyridinealdoxime (336 mg, 2.75 mmol) and then tetramethylguanidine (288 mg, 2.50 mmol). The resulting solution was stirred overnight at rt, evaporated in vacuo and co-evaporated three times from acetonitrile (20 mL). The oily residue was purified on silica using methanol/dichloromethane (1:4) as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (72 mg) as a colorless solid.

^{1}H NMR (DMSO-d_6): δ 3.60 (m, 2H), 3.73 (m, 1H), 4.01 (m, 2H), 5.06 (m, 1H), 5.48 (m, 2H), 6.12 (m, 1H), 6.52 (bs, 2H), 7.70 (s, 1H), 10.75 (bs, 1H).

EXAMPLE 18

2-Amino-5-methyl-7-(β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



Step A 2-Amino-7-(2,3,5-tri-O-benzyl-β-D-arabinofuranosyl)-4-chloro-5-methyl-7H-pyrrolo[2,3-d]pyrimidine

To a solution of 1-O-p-nitrobenzyl-D-arabinofuranose (3.81 g, 6.70 mmol) in DCM was bubbled HBr until TLC (hexane/ethylacetate (2:1)) showed complete reaction (about 30 min). The reaction mixture was filtered and evaporated in vacuo. The oily residue was taken up in acetonitrile (10 mL) and added to a vigorously stirred suspension of 2-amino-4-chloro-5-methyl-7H-pyrrolo[2,3-d]pyrimidine (*Liebigs Ann. Chem.* (1984), 4, 708) (1.11 g, 6.00 mmol) KOH (1.12 g, 20.0 mmol) and tris[2-(2-methoxyethoxy)ethyl]amine (0.216 g, 0.67 mmol) in acetonitrile (80 mL). The resulting suspension was stirred at rt for 30 min, filtered and evaporated in vacuo. The crude product was purified on silica using hexane/ethylacetate (3:1) as the eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (1.13 g) as a colorless foam.

Step B: 2-Amino-7-β-D-arabinofuranosyl-4-chloro-5-methyl-7H-pyrrolo[2,3-d]pyrimidine

To a precooled (−78° C.) solution of the compound from Step A (0.99 g, 1.7 mmol) in dichloromethane (30 mL) was added borontrichloride (1M in dichloromethane) (17 mL, 17.0 mmol) over a 10 min. The resulting solution was stirred at −78° C. for 1 h, allowed to warm to −15° C. and stirred for another 3 h. The reaction was quenched by addition of methanol/dichloromethane (1:1) (15 mL), stirred at −15° C. for 30 min, and pH adjusted to 7.0 by addition of NH_4OH. The mixture was evaporated in vacuo and the resulting oil purified on silica using methanol/dichloromehane (1:9) as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (257 mg) as a colorless foam.

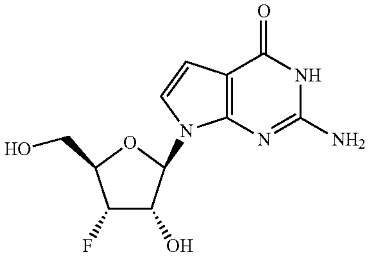
Step C: 2-Amino-7-(β-D-arabinofuranosyl)-5-methyl-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one

To the compound from Step B (157 mg, 0.50 mmol) was added NaOH (2M, aqueous) (2 mL). The resulting solution was stirred at relux for 1 h, cooled and neutralized by addition of HCl (2M, aqueous). The mixture was evaporated in vacuo and the crude product purified on silica using methanol/dichloromehane (2:8) as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (53 mg) as a colorless powder.

^{1}H NMR (DMSO-d$_6$): δ 2.13 (d, 3H), 3.58 (m, 2H), 3.71 (m, 1H), 4.00 (m, 2H), 5.09 (m, 1H), 6.22 (bs, 2H), 5.50 (m, 2H), 6.12 (m, 1H), 6.64 (s, 1H), 10.75 (bs, 1H).

EXAMPLE 19

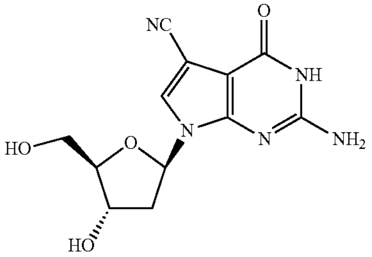
2-Amino-7-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



A solution 1-O-acetyl-2-O-benzyl-5-O-(p-toluoyl)-3-deoxy-3-fluoro-D-ribofuranose (410 mg, 1.01 mmol) (prepared by a modified method described for similar sugar derivatives, *Helv. Chim. Acta* 82: 2052 (1999) and *J. Med. Chem.* 1991, 34, 2195) in anhydrous CH_2Cl_2 (1.5 mL) was cooled to −15° C. in a dry ice/CH_3CN bath. After cooling the reaction mixture for 10 min. under the argon atmosphere, 33% HBr/AcOH (370 μL, 1.5 equiv.) was added slowly over 20 min keeping the bath temperature around −15° C. After the addition was complete, the reaction mixture was stirred at −10° C. for 1 hr. The solvent was removed under reduced pressure and the residue azeotroped with anhydrous toluene (5×10 mL). In a separate flask, 2-amino-4-chloro-7H-pyrrolo[2,3-d]pyrimidine (210 mg, 1.2 mmol) was suspended in anhydrous CH_3CN (10 mL) and cooled to −10° C. To this was added 60% NaH dispersion in oil (57 mg) in two portions, and the reaction mixture was stirred for 45 min. during which time the solid dissolved and the bath temperature rose to 0° C. The bath was removed and stirring was continued for about 20 additional min. It was cooled back to −10° C. and the bromo sugar, prepared above, was taken up in anhydrous CH_3CN (1.5 mL) and added slowly to the anion of nucleobase. After the addition was complete, the reaction mixture was stirred for an additional 45 min allowing the temperature of the reaction to rise to 0° C. The bath was removed and the reaction allowed to stir at room temperature for 3 hr. Methanol was added carefully to the reaction mixture and the separated solid removed by filtration. The solvent was removed under reduced pressure and the residual oil dissolved in EtOAc (50 mL) and washed with water (3×20 mL). The organic layer was dried over Na_2SO_4 and concentrated to give an oil. It was purified by column chromatography to furnish fully protected 2-amino-7-(5-O-(p-toluoyl)-2-O-benzyl-3-deoxy-3-fluoro-β-D-ribofuranosyl)-4-chloro-7H-pyrrolo[2,3-d]pyrimidine (190 mg) as an α/β mixture (1:1). After conversion of 4-chloro to 4-oxo by heating the compound with 2N NaOH/dioxane mixture at 105° C. and after the usual workup the residue was debenzylated using 20 mol % w/w of 10% Pd/C and ammonium formate in refluxing methanol to give title compound after purification by HPLC; yield 10%. ESMS: calcd. for $C_{11}H_{13}FN_4O_4$ 284.24, found 283.0 (M+1).

EXAMPLE 20

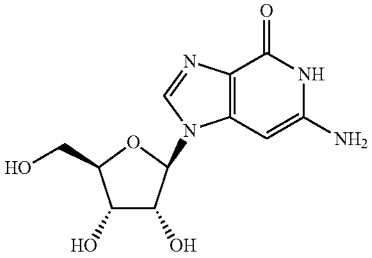
2-Amino-3,4-dihydro-4-oxo-7-(2-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile



This compound was prepared following the procedures described in *Synthesis* 1327 (1998).

EXAMPLE 21

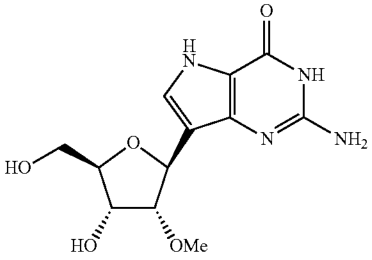
6-Amino-1-(β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one



This compound was prepared following the conditions described in *J. Am. Chem. Soc.* 97: 2916 (1975).

EXAMPLE 22

2-Amino-7-(2-O-methyl-β-D-ribofuranosyl)-5H-pyrrolo[3,2-d]pyrimidin-4-(3H)-one



To a suspension of 2-amino-5H-pyrrolo[3,2-d]pyrimidin-4(3H)-one (9-deazaguanine) (0.454 g, 3.0 mmol) (prepared according to *J. Org. Chem.* 1978, 43, 2536) and 2-O-methyl-

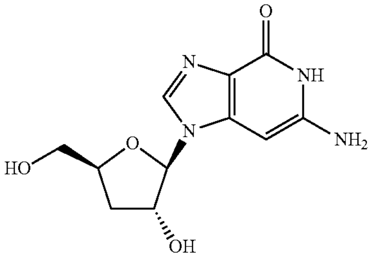
1,3,5-tri-O-benzoyl-β-D-ribofuranose (1.54 g, 3.2 mmol) in dry nitromethane (23 mL) at 60° C. was added stannic chloride (0.54 mL, 4.5 mmol). The reaction mixture was maintained at this temperature for 0.5 hr., cooled and poured onto ice-cold saturated sodium bicarbonate solution (70 mL). The insoluble material was filtered through florisil and washed with ethyl acetate (3×50 mL). The filtrate was extracted with ethyl acetate (2×50 mL), and organic layer was washed with water (2×50 mL), dried over Na_2SO_4 and evaporated to dryness. Chromatography of the resulting foam on silica gel with CH_2Cl_2/MeOH (14:1) afforded the benzoylated product (0.419 g, 30% yield). To a suspension of the benzoylated product (0.25 g) in MeOH (2.4 mL) was added t-butylamine (0.52 mL) and stirring at room temperature was continued for 24 hrs. followed by addition of more t-butylamine (0.2 mL). The reaction mixture was stirred at ambient temperature overnight, concentrated in vacuum and the residue was purified by flash chromatography over silica gel using CH_2Cl_2/MeOH (85:15) as eluent giving the desired compound as a foam (0.80 g).

^1H NMR (200 MHz, DMSO-d_6): δ Hz 3.28 (s, 3H), 3.40-3.52 (m, 3H), 3.87-3.90 (m, 1H), 4.08-4.09 (m, 1H), 4.67 (d, 1H, J=5.2 Hz), 4.74 (d, 1H, J=7.0 Hz), 5.62 and 5.50 (2 bs, 3H), 7.14 (d, 1H, J=2.6 Hz), 10.43 (s, 1H), 11.38 (s, 1H);

Mass spectrum: calcd. for $C_{12}H_{16}N_4O_5$: 296.28; found: 295.11.

EXAMPLE 23

6-Amino-1-(3-deoxy-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridine-4(5H)-one (3'-deoxy-3-deazaguanosine)



Step A: 3-Deoxy-4-O-p-toluoyl-2-O-acetyl-β-D-ribofuranosyl acetate

A solution of 3-deoxy-4-O-p-toluoyl-1,2-O-isopropylidene-β-D-ribofuranose (*Nucleosides Nucleotides* 1994, 13, 1425 and *Nucleosides Nucleotides* 1992, 11, 787) (5.85 g, 20 mmol) in 64 mL of 80% acetic acid was stirred at 85° C. overnight. The reaction mixture was concentrated and co-evaporated with toluene. The residue was dissolved in 90 mL of pyridine. Acetic anhydride (6 mL) was added at 0° C., and the reaction mixture was stirred at rt for 6 h. After condensation, the residue was dissolved in ethyl acetate and washed with aqueous sodium bicarbonate solution, water and brine. The organic phase was dried and concentrated. Chromatographic purification on a silica gel column using 3:1 and 2:1 hexanes-EtOAc as eluent provided 5.51 g of the title compound as a clear oil.

^1H NMR ($CDCl_3$): δ 1.98 (s, 3H), 2.09 (s, 3H), 2.15-2.35 (m, 2H), 2.41 (s, 3H), 4.27-4.42 (m, 1H), 4.46-4.58 (m, 1H), 4.65-4.80 (m, 1H), 5.21-5.28 (m, 1H), 6.20 (s, 1H), 7.19-7.31 (m, 2H), 7.90-8.01 (m, 2H).

Step B: Methyl 5-cyanomethyl-1-(3-deoxy-4-O-p-toluoyl-2-O-acetyl-β-D-ribofuranosyl)-1H-imidazole-4-carboxylate

A mixture of methyl 5(4)-(cyanomethyl)-1H-imidazole-4(5)-carboxylate (*J. Am. Chem. Soc.* 1976, 98, 1492 and *J. Org. Chem.* 1963, 28, 3041) (1.41 g, 8.53 mmol), 1,1,1,3,3,3-hexamethyldisilazane (20.5 mL) and ammonium sulfate (41 mg) was refluxed at 125° C. under Ar atmosphere for 18 h. After evaporation, the residue was dissolved in 10 mL of dichloroethane. A solution of the compound from Step A (2.86 g, 8.5 mmol) in 10 mL of dichloroethane was added followed by addition of $SnCl_4$ (1.44 mL, 3.20 g). The resulted reaction mixture was stirred at rt overnight and diluted with chloroform. The mixture was washed with aqueous sodium bicarbonate, water and brine. The organic phase was dried and concentrated. Chromatographic purification of the residue on a silica gel column using 1:1, 1:2, and 1:3 hexanes-EtOAc as eluent provided 2.06 g of the title compound as a white foam.

^1H NMR ($CDCl_3$): δ 2.15 (s, 3H), 2.28-2.40 (m, 2H), 2.38 (s, 3H), 3.87 (s, 3H), 4.46 (dd, 2H, J=7.6, 2.0 Hz), 4.50-4.57 (m, 1H), 4.68-4.75 (m, 1H), 4.76-4.83 (m, 1H), 5.41 (d, 1H, J=5.6 Hz), 5.91 (s, 1H), 7.24-7.28 (m, 2H), 7.80 (s, 1H), 7.82-7.90 (m, 2H); ^{13}C NMR ($CDCl_3$) δ 13.1, 20.7, 21.6, 31.5, 51.8, 63.5, 77.9, 79.2, 89.8, 115.1, 126.2, 129.3, 129.5, 131.7, 135.1, 144.3, 163.1, 166.1, 170.3.

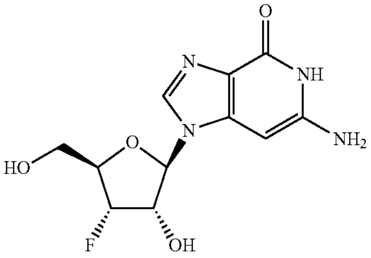
Step C: 6-Amino-1-(3-deoxy-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridine-4(5H)-one

A solution of the compound from Step B (2.00 g, 4.53 mmol) in methanol (30 mL) was saturated with ammonia at 0° C. Concentrated ammonium hydroxide (30 mL) was added and the sealed metal reactor was heated at 85° C. for 5 h. After cooling to rt, the reaction mixture was transferred directly onto a silica gel column. Elution with 4:1, 3:1 and 2:1 $CHCl_3$-MeOH provided 0.79 g of the title compound as a white solid.

^1H NMR (DMSO-d_6): δ 2.41-2.46 (m, 1H), 2.52-2.58 (m, 1H), 3.48-3.55 (m, 1H), 3.60-3.70 (m, 1H), 4.27-4.36 (m, 2H), 4.97 (t, 1H, J=5.6 Hz), 5.44 (s, 1H), 5.47 (s, 1H), 5.60 (s, 2H), 5.66, (d, 1H, J=4.4 Hz), 7.90 (s, 1H), 10.33 (s, 1H); ^{13}C NMR (DMSO d_6) δ 34.1, 62.4, 70.4, 74.7, 80.4, 91.6, 123.0, 136.3, 141.9, 147.6, 156.5.

EXAMPLE 24

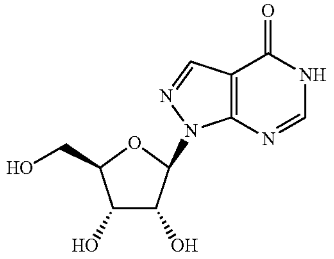
6-Amino-1-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(3H)-one



This compound was prepared in a manner similar to the preparation of 2-amino-7-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one (Example 23).

EXAMPLE 25

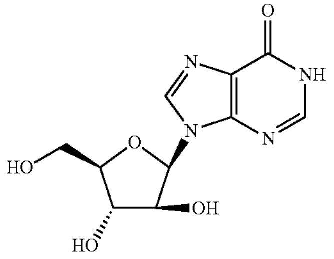
1-(β-D-Ribofuranosyl)-1H-pyrazolo[3,4-d]pyrimidin-4(3H)-one (Allopurinol riboside)



This compound was obtained from commercial sources.

EXAMPLE 26

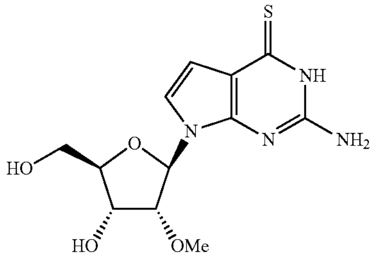
9-(β-D-Arabinofuranosyl)-9H-purin-6(1H)-one



This compound was prepared following the conditions described in *J. Med. Chem.* 18: 721 (1975).

EXAMPLE 27

2-Amino-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-thione

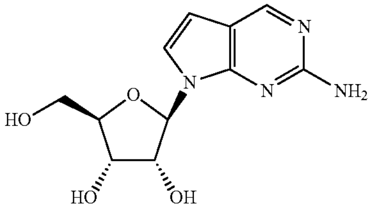


A solution of the compound from Example 11, Step C (1.5 g, 5 mmol), thiourea (0.4 g, 5.2 mmol.) in abs. EtOH was refluxed for 16 hrs. The solution was evaporated and the resulting oil chromatographed on silica gel (EtOAc/MeOH: 9/1) to afford the desired product as a foam.

^1H NMR (DMSO-d_6): δ 3.30 (s, 3H), 5.00-5.06 (t, 1H), 5.19 (d, 1H), 5.95 (d, 1H), 6.43 (d, 1H), (d, 1H).

EXAMPLE 28

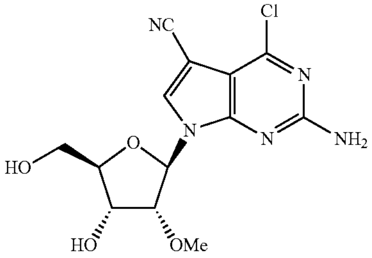
2-Amino-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



This compound was obtained from commercial sources.

EXAMPLE 29

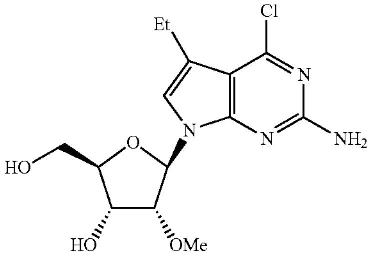
2-Amino-4-chloro-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile



This compound was prepared as described in Example 13, Steps A-C.

EXAMPLE 30

2-Amino-4-chloro-5-ethyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



Step A: 2-Amino-4-chloro-5-ethyl-7-[3,5-O-(tetraisopropyldisiloxane-1,3-diyl)-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine

To a solution of 2-amino-4-chloro-5-ethyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine (0.300 g, 0.913

mmol) in pyridine (8 mL) was added 1,3-dichloro-1,1,3,3-tetraisopropyldisiloxane (0.317 g, 1.003 mmol) dropwise. The solution stirred at rt overnight, evaporated in vacuo to an oil, and evaporated repeatedly from acetonitrile. The crude product was purified on silica using 5% methanol in dichloromethane as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (254 mg) as a colorless solid.

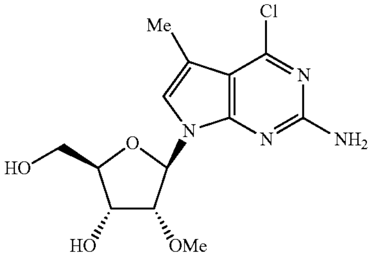
Step B: 2-Amino-4-chloro-5-ethyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

To a pre-cooled solution (0° C.) of the compound from step A (192 mg, 0.337 mmol) in DMF (3 mL) was added methyl iodide (45.4 mg, 0.320 mmol) and then NaH (60% in mineral oil) (8.10 mg, 0.320 mmol). The mixture was stirred at rt for 45 minutes and then poured into a stirred mixture of saturated aqueous ammonium chloride (10 mL) and ethyl acetate (10 mL). The organic phase phase was washed with brine (10 mL) and dried over $MgSO_4$ and evaporated in vacuo. The resulting oily residue was taken up in THF (5 mL) and tetrabutylammonium fluoride (1.1 mmol/g on silica) (0.529 g, 0.582 mmol) was added. The mixture was stirred for 30 minutes, filtered and the filtrate evaporated in vacuo. The crude product was purified on silica using 10% methanol in dichloromethane as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (66 mg) as a colorless solid.

^1H NMR (DMSO-d_6): δ 1.15 (t, 3H), 2.65 (q, 2H), 3.20 (s, 3H), 3.51 (m, 2H), 3.84 (m, 1H), 4.04 (m, 1H), 4.21 (m, 1H), 4.99 (m, 2H), 5.15 (m, 2H), 6.07 (m, 2H), 6.62 (s br, 2H), 7.06 (s, 2H).

EXAMPLE 31

2-Amino-4-chloro-5-methyl-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

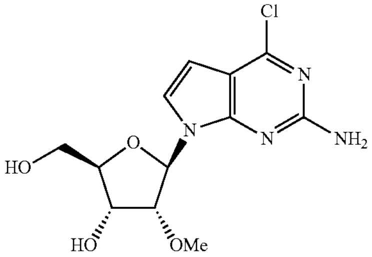


This compound was prepared as described in Example 14, Step A.

^1H NMR (CD_3OD): δ 2.33 (s, 3H), 3.39 (s, 1H), 3.72, 3.83 (2dd, 2H), 4.03 (m, 1H), 4.17 (t, 1H), 4.39 (dd, 1H), 5.98 (d, 1H, J=5.9 Hz), 6.7 (bs, 2H), 7.01 (s, 1H).

EXAMPLE 32

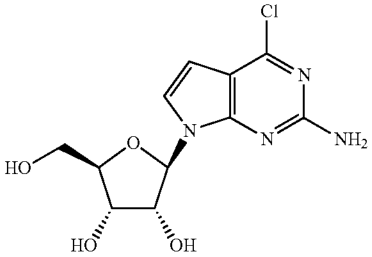
2-Amino-4-chloro-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



This compound was synthesized as described in Example 11, Steps A-C.

EXAMPLE 33

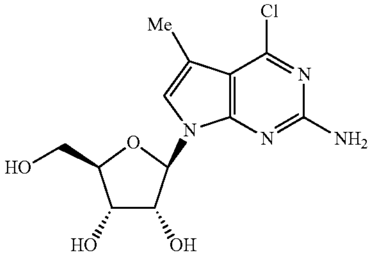
2-Amino-4-chloro-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



This compound was prepared following the procedures described in *Helv. Chim. Acta* 73: 1879 (1990).

EXAMPLE 34

2-Amino-4-chloro-5-methyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

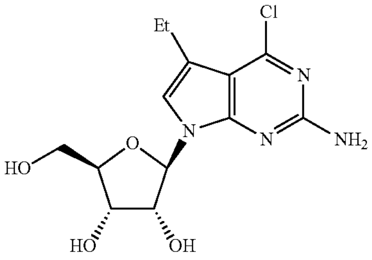


The compound was prepared as described in Example 12, Steps A-B.

^{1}H NMR (DMSO-d_6): δ 2.29 (s, 3H), 3.54 (m, 2H), 3.84 (m, 1H), 4.04 (dd, 1H, J_1=3.0, J_2=4.9 Hz), 4.80-5.50 (bs, 3H), 4.28 (t, 1H), 5.98 (d, 1H, J=6.5 Hz), 6.7 (bs, 2H), 7.13 (s, 1H).

EXAMPLE 35

2-Amino-4-chloro-5-ethyl-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

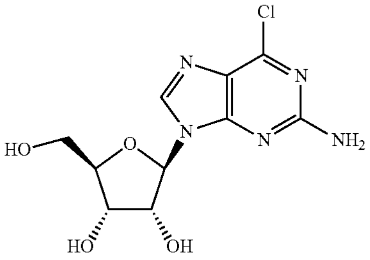


This compound was prepared as described in Example 9, Steps A-B.

^{1}H NMR (DMSO-d_6): δ 2.00 (t, 3H), 2.69 (q, 2H), 3.48 (dd, 1H, J_1=4.2 Hz, J_2=11.8 Hz), 3.56 (dd, 1H, J_1=4.3 Hz, J_2=11.8 Hz), 3.80 (m, 1H), 4.02 (dd, 1H, J_1=3.1 Hz, J_2=5.0 Hz), 4.62 (t, 1H), 5.0 (bs, 2H), 5.2 (bs, 1H), 5.60 (d, 1H, J=6.4 Hz), 6.61 (bs, 2H), 7.09 (s, 1H).

EXAMPLE 36

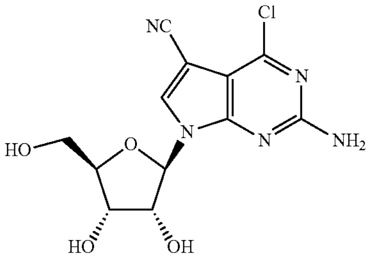
2-Amino-6-chloro-9-(β-D-ribofuranosyl)-9H-purine



This compound was obtained from commercial sources.

EXAMPLE 37

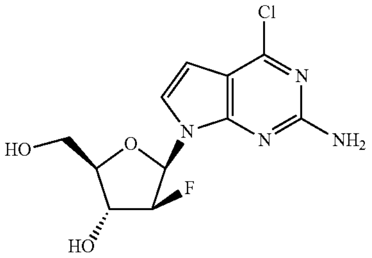
2-Amino-4-chloro-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile



This compound was prepared following the procedures described in *J. Chem. Soc. Perkin Trans.* 1, 2375 (1989).

EXAMPLE 38

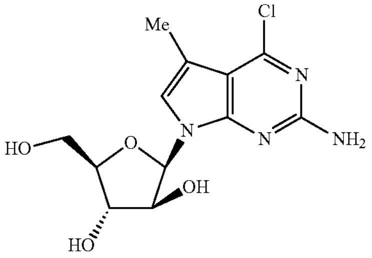
2-Amino-4-chloro-7-(2-deoxy-2-fluoro-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



This compound was prepared following the procedures described in *J. Med. Chem.* 38: 3957 (1995).

EXAMPLE 39

2-Amino-4-chloro-5-methyl-7-(β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

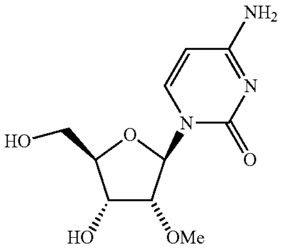


The compound was prepared as described in Example 18, Steps A-B.

^{1}H NMR (DMSO-d$_6$): δ 2.24 (s, 3H), 3.60 (m, 3H), 3.98 (m, 2H), 4.98 (m, 1H), 5.43 (bs, 2H), 6.25 (s, 1H), 6.57 (bs, 2H), 7.01 (s, 1H).

EXAMPLE 40

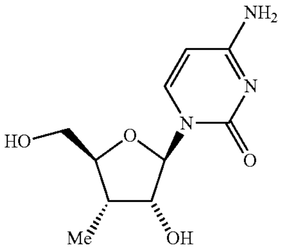
2'-O-Methylcytidine



This compound was obtained from commercial sources.

EXAMPLE 41

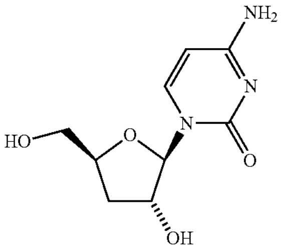
3'-Deoxy-3'-methylcytidine



This compound was prepared following the procedures described in U.S. Pat. No. 3,654,262 (1972), which is incorporated by reference herein in its entirety.

EXAMPLE 42

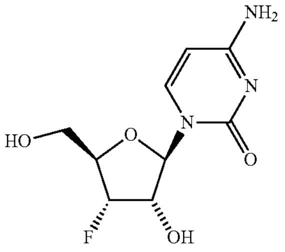
3'-Deoxycytidine



This compound was obtained from commercial sources.

EXAMPLE 43

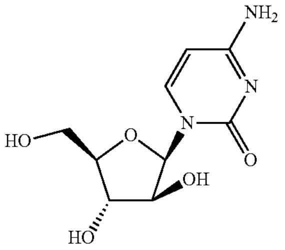
3'-Deoxy-3'-fluorocytidine



This compound was prepared following the procedures described in *J. Med. Chem.* 34: 2195 (1991).

EXAMPLE 44

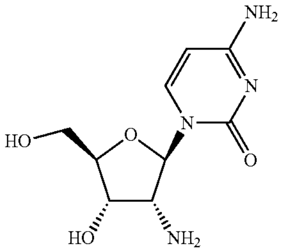
1-(β-D-Arabinofuranosyl)-1H-cytosine



This compound was obtained from commercial sources.

EXAMPLE 45

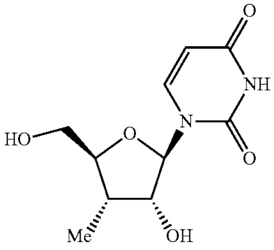
2'-Amino-2'-deoxycytidine



This compound was obtained from commercial sources.

EXAMPLE 46

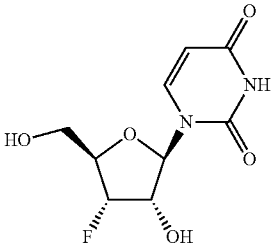
3'-Deoxy-3'-methyluridine



This compound was prepared following procedures described in U.S. Pat. No. 3,654,262, which is incorporated by reference herein in its entirety.

EXAMPLE 47

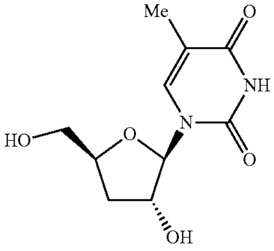
3'-Deoxy-3'-fluorouridine



This compound was prepared following procedures described in *J. Med. Chem.* 34: 2195 (1991) and *FEBS Lett.* 250: 139 (1989).

EXAMPLE 48

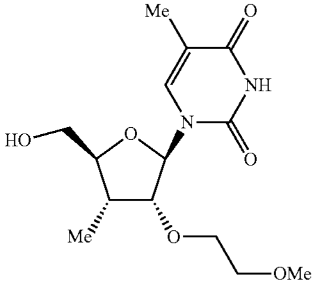
3'-Deoxy-5-methyluridine



This compound was obtained from commercial sources.

EXAMPLE 49

3'-Deoxy-2'-O-(2-methoxyethyl)-3'-methyl-5-methyluridine



Step A: 5'-O-(tert-butyldiphenylsilyl)-3'-O-(3-tert-butylphenoxythiocarbonyl)-2'-O-(2-methoxyethyl)-5-methyluridine

This compound was synthesized by the reaction of the corresponding 5'-protected-2'-substituted-5-methyluridine with 3'-t-butylphenoxy chlorothionoformate following the similar procedure for the preparation of 3'-phenoxythiocarbonyl-2'-deoxy derivative (*Synthesis* 1994, 1163).

Step B: 5'-O-(tert-Butyldiphenylsilyl)-3'-deoxy-2'-O-(2-methoxyethyl)-3'-(2-phenylethenyl)-5-methyluridine

To a solution of 5'-O-(tert-butyldiphenylsilyl)-3'-O-(3-tert-butylphenoxythiocarbonyl)-2'-O-(2-methoxyethyl)-5-methyluridine (15.0 g, 20.0 mmol) in 150 mL of benzene was added PhCH═CHSnBu$_3$ (18.7 g, 50 mmol). The resulting solution was degassed three times with argon at rt and 45° C. After AIBN (1.0 g, 6.1 mmol) was added, the resulting solution was refluxed for 2 h. Another portion of AIBN (1.0 g, 6.1 mmol) was added after cooling to about 40° C. and refluxed for 2 h. This procedure was repeated until the starting material disappeared. The solvent was evaporated and the residue was purified by flash chromatography on a silica gel column using 10:1 and 5:1 hexanes-EtOAc as eluent to give 1.74 g of 5'-O-(tert-butyldiphenylsilyl)-3'-deoxy-2'-O-(2-methoxyethyl)-3'-(2-phenylethenyl)-5-methyluridine as a white foam. ^{1}H NMR (CDCl$_3$): δ 1.13, (s, 9H), 1.43 (s, 3H), 3.18-3.30 (m, 1H), 3.37 (s, 3H), 3.58-3.62 (m, 2H), 3.79-3.80 (m, 2H), 4.06-4.37 (m, 4H), 4.95 (s, 1H), 6.25-6.40 (m, 1H), 6.62 (d, 1H, J=16 Hz), 7.27-7.71 (m, 16H), 9.21 (s, 1H); ^{13}C NMR (CDCl$_3$) δ 11.9, 19.6, 27.2, 45.3, 59.0, 62.1, 70.2, 72.0, 84.6, 87.1, 90.2, 110.4, 122.8, 126.4, 127.8, 128.0, 128.3, 128.6, 130.0, 132.7, 133.5, 134.7, 135.3, 135.4, 136.9, 150.3, 154.1; HRMS (FAB) m/z 641.302 (M+H)$^+$ ($C_{37}H_{45}N_2O_6Si$ requires 641.304).

Step C: 5'-O-(tert-Butyldiphenylsilyl)-3'-deoxy-3'-(hydroxymethyl)-2'-O-(2-methoxyethyl)-5-methyluridine

To a solution of 5'-O-(tert-butyldiphenylsilyl)-3'-deoxy-2'-O-(2-methoxyethyl)-3'-(2-phenylethenyl)-5-methyluridine. (5.0 g, 7.8 mmol) and N-methylmorpholine N-oxide (NMO)

(1.47 g, 12.5 mmol) in 150 mL of dioxane was added a catalytic amount of osmium tetraoxide (4% aqueous solution, 2.12 mL, 85 mg, 0.33 mmol). The flask was covered by aluminum foil and the reaction mixture was stirred at rt overnight. A solution of $NaIO_4$ (5.35 g, 25 mmol) in 5 mL of water was added to the above stirred reaction mixture. The resulting reaction mixture was stirred for 1 h at 0° C. and 2 h at rt, followed by addition of 10 mL of ethyl acetate. The mixture was filtered through a celite pad and washed with ethyl acetate. The filtrate was washed 3 times with 10% aqueous $Na_2S_2O_3$ solution until the color of aqueous phase disappeared. The organic phase was further washed with water and brine, dried (Na_2SO_4) and concentrated. The aldehyde thus obtained was dissolved in 130 mL of ethanol-water (4:1, v/v). Sodium borohydride ($NaBH_4$) (1.58 g, 40 mmol) was added in portions at 0° C. The resulting reaction mixture was stirred at rt for 2 h and then treated with 200 g of ice water. The mixture was extracted with ethyl acetate. The organic phase was washed with water and brine, dried (Na_2SO_4) and concentrated. The resulted residue was purified by flash chromatography on a silica gel column using 2:1, 1:1 and 1:2 hexanes-EtOAc as eluents to give 1.6 g of 5'-O-(tert-butyldiphenylsilyl)-3'-deoxy-3'-(hydroxymethyl)-2'-O-(2-methoxyethyl)-5-methyluridine as a white foam.

^1H NMR ($CDCl_3$): δ 1.09 (s, 9H), 1.50 (s, 3H), 2.25 (bs, 1H), 2.52-2.78 (m, 1H), 3.38 (s, 3H), 3.52-4.25 (m, 10H), 5.86 (s, 1H), 7.38-7.70 (m, 11H), 9.95 (bs, 1H); ^{13}C NMR ($CDCl_3$): δ 12.1, 19.5, 27.1, 43.1, 58.2, 58.8, 63.1, 69.5, 71.6, 82.3, 86.1, 89.8, 110.5, 128.0, 130.2, 132.5, 133.2, 135.1, 135.3, 136.5, 150.5, 164.4; HRMS (FAB) m/z 569.268 $(M+H)^+$ ($C_{30}H_{41}N_2O_7Si$ requires 569.268).

Step D: 5'-O-(tert-Butyldiphenylsilyl)-3'-deoxy-3'-(iodomethyl)-2'-O-(2-methoxyethyl)-5-methyluridine

To a solution of 5'-O-(tert-butyldiphenylsilyl)-3'-deoxy-3'-(hydroxymethyl)-2'-O-(2-methoxyethyl)-5-methyluridine (1.34 g, 2.35 mmol) in 25 mL of anhydrous DMF under stirring was added sequentially at 0° C. 2,6-lutidine (0.55 mL, 0.51 g, 4.7 mmol, 2.0 equiv) and methyl triphenoxy-phosphonium iodide (1.28 g, 2.83 mmol). The resulting reaction mixture was stirred at 0° C. for 1 h and at rt for 2 h. The reaction mixture was diluted with 10 mL of ethyl acetate and washed twice with 0.1 N $Na_2S_2O_3$ aqueous solution to remove iodine. The organic phase was further washed with aqueous $NaHCO_3$ solution, water, and brine. The aqueous phases were back extracted with ethyl acetate. The combined organic phases were dried (Na_2SO_4) and concentrated. The resulting residue was purified by flash chromatography on a silica gel column using 5:1, 3:1 and then 1:1 hexanes-EtOAc to provide 1.24 g of 5'-O-(tert-butyldiphenylsilyl)-3'-deoxy-3'-(iodomethyl)-2'-O-(2-methoxyethyl)-5-methyluridine as a white foam.

^1H NMR ($CDCl_3$): δ 1.13 (s, 9H), 1.62 (s, 3H), 2.64-2.85 (m, 2H), 3.20-3.35 (m, 1H), 3.38 (s, 3H), 3.50-4.25 (m, 8H), 5.91 (s, 1H), 7.32-7.50 (m, 6H), 7.60 (s, 1H), 7.62-7.78 (m, 4H), 10.46 (s, 1H); ^{13}C NMR ($CDCl_3$): δ 12.4, 19.5, 27.2, 45.0, 58.0, 62.5, 70.3, 71.9, 83.3, 85.6, 88.9, 110.5, 128.1, 128.2, 130.1, 130.3, 132.4, 132.9, 135.0, 135.4, 135.6, 150.7, 164.7; HRMS (FAB) m/z 679.172 $(M+H)^+$ ($C_{30}H_{40}IN_2O_6Si$ requires 679.170).

Step E: 3'-Deoxy-3'-(iodomethyl)-2'-O-(2-methoxyethyl)-5-methyluridine

A solution of 5'-O-(tert-butyldiphenylsilyl)-3'-deoxy-3'-(iodomethyl)-2'-O-(2-methoxyethyl)-5-methyluridine (1.12 g, 1.65 mmol) and triethylamine trihydrofluoride (1.1 mL, 1.1 g, 6.7 mmol) in 20 mL of THF was stirred at rt for 24 h. The reaction mixture was diluted with 50 mL of ethyl acetate and washed with water and brine. The organic phase was dried (Na_2SO_4) and concentrated. The residue was purified by flash chromatography on a silica gel column. Gradient elution with 2:1, 1:2 and then 1:3 hexanes-EtOAc provided 504 mg of the title compound as a white foam.

^1H NMR (CD_3OD): δ 1.87 (s, 3H), 2.47-2.75 (m, 1H), 3.18-3.37 (m, 2H), 3.40 (s, 3H), 3.59-3.70 (m, 2H), 3.71-3.90 (m, 2H), 3.92-4.17 (m, 4H), 5.87 (s, 1H), 8.17 (s, 1H); ^{13}C NMR (CD_3OD): δ 12.5, 45.2, 59.2, 60.9, 71.0, 72.9, 85.4, 87.3, 89.7, 110.5, 138.0, 152.1, 166.6; HRMS (FAB) m/z 441.053 $(M+H)^+$ ($C_{14}H_{22}IN_2O_6$ requires 441.052).

Step F: 3'-Deoxy-5'-O-(4-methoxytrityl)-3'-(iodomethyl)-2'-O-(2-methoxyethyl)-5-methyluridine

A mixture of 3'-deoxy-3'-(iodomethyl)-2'-O-(2-methoxyethyl)-5-methyluridine (472 mg, 1.1 mmol), diisopropylethylamine (0.79 mL, 0.586 g, 4.5 mmol), and p-anisyl chlorodiphenyl methane (4'-methoxytrityl chloride, MMT-Cl) (1.32 g, 4.27 mmol) in 6 mL of ethyl acetate and 4 mL of THF was stirred at rt for 48 h. The reaction mixture was diluted with ethyl acetate and washed with water, followed by brine. The organic phase was dried (Na_2SO_4) and concentrated. The crude product was purified by flash chromatography on a silica gel column. Gradient elution with 3:1, 2:1, 1:1, and then 1:3 hexanes-EtOAc provided 690 mg of the title compound as a white foam.

^1H NMR ($CDCl_3$): δ 1.46 (s, 3H), 2.70-2.89 (m, 2H), 3.19-3.31 (m, 2H), 3.39 (s, 3H), 3.58-3.70 (m, 3H), 3.80 (s, 3H), 3.80-3.94 (m, 1H), 4.05-4.25 (m, 3H), 5.89 (s, 1H), 6.85 (s, 1H), 6.89 (s, 1H), 7.24-7.48 (m, 12H), 7.78 (s, 1H), 9.69 (s, 1H); ^{13}C NMR ($CDCl_3$): δ 12.3, 45.3, 55.3, 58.9, 61.6, 70.2, 71.9, 82.6, 85.6, 87.1, 89.1, 110.5, 113.4, 127.4, 128.2, 128.4, 130.5, 134.7, 135.3, 143.6, 143.7, 150.5, 158.9, 164.6. HRMS (FAB) m/z 735.155 $(M+Na)^+$ ($C_{34}H_{37}IN_2O_7Na$ requires 735.154).

Step G: 3'-Deoxy-5'-O-(4-methoxytrityl)-3'-methyl-2'-O-(2-methoxyethyl)-5-methyluridine

A mixture of ammonium phosphinate (410 mg, 5.1 mmol) and 1,1,1,3,3,3-hexamethyldisilazane (1.18 mL, 0.90 g, 5.59 mmol) was heated at 100-110° C. for 2 h under nitrogen atmosphere with condenser. The intermediate BTSP(bis[trimethylsilyl]phosphinate) was cooled to 0° C. and 5 mL of dichloromethane was injected. To this mixture was injected a solution of 3'-deoxy-5'-O-(4-methoxytrityl)-3'-(iodomethyl)-2'-O-(2-methoxyethyl)-5-methyluridine (0.78 g, 1.1 mmol) and diisopropylethylamine (0.39 mL, 287 mg, 2.23 mmol) in 7 mL of dichloromethane. After the reaction mixture was stirred at rt overnight, a mixture of THF-MeOH-NEt_3 (3/6/0.3 mL) was added and continued to stir for 1 h. The reaction mixture was filtered through a pad of celite and washed with dichloromethane. The solvent was evaporated and the residue was purified by flash chromatography on a silica gel column using 2:1, 1:1, and then 1:2 hexanes-EtOAc as eluent providing 380 mg of the title compound.

^1H NMR ($CDCl_3$): δ 0.97 (d, 3H, J=6.8 Hz), 1.41 (s, 3H), 2.35-2.55 (m, 1H), 3.27 (dd, 1H, J=11.0, 3.0 Hz), 3.37 (s, 3H), 3.54-3.68 (m, 3H), 3.79 (s, 3H), 3.75-3.87 (m, 1H), 3.94 (d, 1H, J=5.0 Hz), 4.03-4.16 (m, 2H), 5.84 (s, 1H), 6.83 (s, 1H), 6.87 (s, 1H), 7.20-7.37 (m, 8H), 7.39-7.50 (m, 4H), 7.86 (s, 1H), 9.50 (s, 1H); ^{13}C NMR ($CDCl_3$): δ 8.7, 12.1, 35.6, 55.3, 59.0, 61.7, 69.8, 72.1, 85.4, 86.4, 86.7, 89.8, 110.0, 113.3, 127.2, 128.0, 128.4, 130.4, 135.0, 135.7, 143.9, 150.5, 158.8, 164.6.

HRMS (FAB) m/z 609.256 $(M+Na)^+$ ($C_{34}H_{38}N_2O_7Na$ requires 609.257).

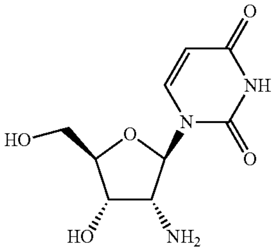
Step H: 3'-Deoxy-3'-methyl-2'-O-(2-methoxyethyl)-5-methyluridine

Trifluoroacetic acid (1.5 mL) was added dropwise to a stirred solution of 3'-deoxy-5'-O-(4-methoxytrityl)-3'-methyl-2'-O-(2-methoxyethyl)-5-methyluridine (370 mg, 0.63 mmol) in 50 mL of chloroform at 0° C. The mixture was stirred at rt for 30 min, concentrated, and then dissolved in ethyl acetate. The solution was washed with dilute sodium bicarbonate and brine. The organic phase was dried (Na_2SO_4) and concentrated. The resulting residue was purified by flash chromatography on a silica gel column. Elution with 1:1, 1:3 and then 0:1 hexanes-EtOAc provided 170 mg of the title compound as a white foam.

^1H NMR ($CDCl_3$): δ 1.03 (d, 3H, J=6.8 Hz), 1.83 (s, 3H), 2.20-2.40 (m, 1H), 3.10-3.28 (m, 1H), 3.35 (s, 3H), 3.50-4.15 (m, 10H), 5.81 (s, 1H), 7.89 (s, 1H), 9.77 (s, 1H); ^{13}C NMR ($CDCl_3$): δ 8.9, 12.4, 34.7, 59.0, 60.6, 69.7, 72.0, 86.3, 89.8, 109.7, 136.9, 150.4, 164.7. HRMS (FAB) m/z 315.154 $(M+H)^+$ ($C_{14}H_{23}N_2O_6$ requires 315.155).

EXAMPLE 50

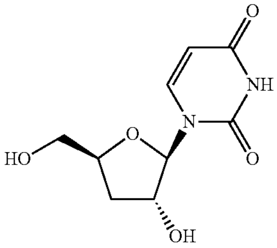
2'-Amino-2'-deoxyuridine



This compound was prepared following the procedures described in *J. Org. Chem.* 61: 781 (1996).

EXAMPLE 51

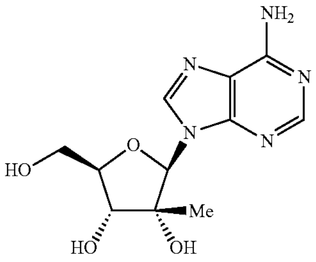
3'-Deoxyuridine



This compound was obtained from commercial sources.

EXAMPLE 52

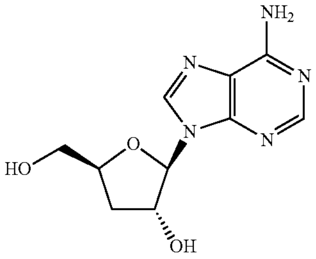
2'-C-Methyladenosine



This compound was prepared following the conditions described in *J. Med. Chem.* 41: 1708 (1998).

EXAMPLE 53

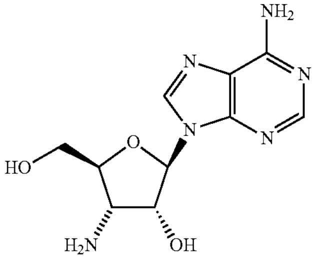
3'-Deoxyadenosine (Cordycepin)



This compound was obtained from commercial sources.

EXAMPLE 54

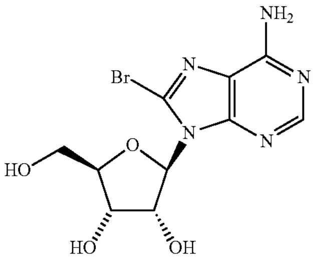
3'-Amino-3'-deoxyadenosine



This compound was prepared following the conditions described in *Tetrahedron Lett.* 30: 2329 (1989).

EXAMPLE 55

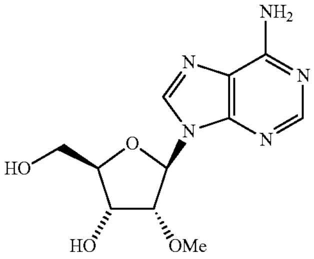
8-Bromoadenosine



This compound was obtained from commercial sources.

EXAMPLE 56

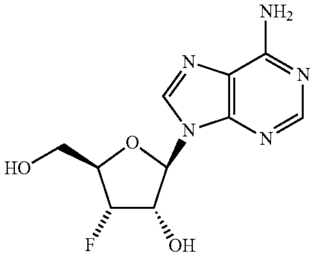
2'-O-Methyladenosine



This compound was obtained from commercial sources.

EXAMPLE 57

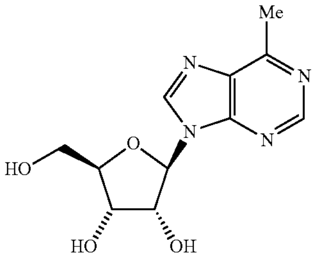
3'-Deoxy-3'-fluoroadenosine



This compound was prepared following the procedures described in *J. Med. Chem.* 34: 2195 (1991).

EXAMPLE 58

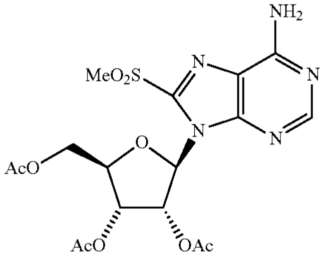
6-Methyl-9-(β-D-ribofuranosyl)-9H-purine



This compound was prepared following the procedures described in *Nucleosides, Nucleotides, Nucleic Acids* 19: 1123 (2000).

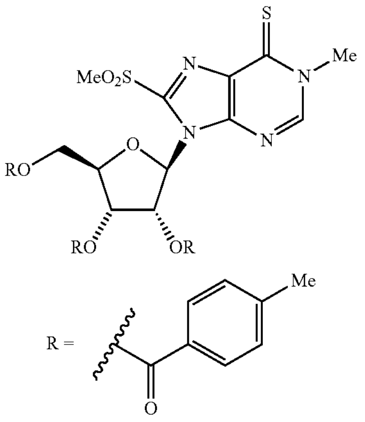
EXAMPLE 59

2',3',5'-tri-O-acetyl-8-methylsulfonyladenosine



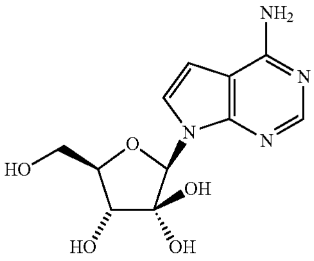
EXAMPLE 60

1-Methyl-9-[2,3,5-tri-O-(p-toluoyl)-β-D-ribofuranosyl]-9H-purine-6(1H)-thione



EXAMPLE 61

4-Amino-7-(2-C-methyl-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

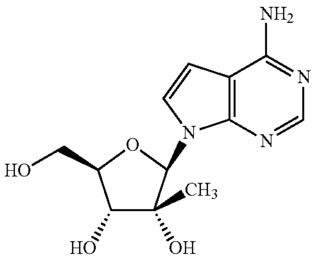


To chromium trioxide (1.57 g, 1.57 mmol) in dichloromethane (DCM) (10 mL) at 0° C. was added acetic anhydride (145 mg, 1.41 mmol) and then pyridine (245 mg, 3.10 mmol). The mixture was stirred for 15 min, then a solution of 7-[3,5-O-[1,1,3,3-tetrakis(1-methylethyl)-1,3-disiloxanediyl]-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidin-4-amine [for preparation, see *J. Am. Chem. Soc.* 105: 4059 (1983)] (508 mg, 1.00 mmol) in DCM (3 mL) was added. The resulting solution was stirred for 2 h and then poured into ethyl acetate (10 mL), and subsequently filtered through silica gel using ethyl acetate as the eluent. The combined filtrates were evaporated in vacuo, taken up in diethyl ether/THF (1:1) (20 mL), cooled to −78° C. and methylmagnesium bromide (3M, in THF) (3.30 mL, 10 mmol) was added dropwise. The mixture was stirred at −78° C. for 10 min, then allowed to come to room temperature (rt) and quenched by addition of saturated aqueous ammonium chloride (10 mL) and extracted with DCM (20 mL). The organic phase was evaporated in vacuo and the crude product purified on silica gel using 5% methanol in dichloromethane as eluent. Fractions containing the product were pooled and evaporated in vacuo. The resulting oil was taken up in THF (5 mL) and tetrabutylammonium fluoride (TBAF) on silica (1.1 mmol/g on silica) (156 mg) was added. The mixture was stirred at rt for 30 min, filtered, and evaporated in vacuo. The crude product was purified on silica gel using 10% methanol in dichloromethane as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired compound (49 mg) as a colorless solid.

^{1}H NMR (DMSO-d_6): δ 1.08 (s, 3H), 3.67 (m, 2H), 3.74 (m, 1H), 3.83 (m, 1H), 5.19 (m, 1H), 5.23 (m, 1H), 5.48 (m, 1H), 6.08 (1H, s), 6.50 (m, 1H), 6.93 (bs, 2H), 7.33 (m, 1H), 8.02 (s, 1H).

EXAMPLE 62

4-Amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



Step A: 3,5-Bis-O-(2,4-dichlorophenylmethyl)-1-O-methyl-α-D-ribofuranose

A mixture of 2-O-acetyl-3,5-bis-O-(2,4-dichlorophenylmethyl)-1-O-methyl-α-D-ribofuranose [for preparation, see: *Helv. Chim. Acta* 78: 486 (1995)] (52.4 g, 0.10 mol) in methanolic K_2CO_3 (500 mL, saturated at room temperature) was stirred at room temperature for 45 min. and then concentrated under reduced pressure. The oily residue was suspended in CH_2Cl_2 (500 mL), washed with water (300 mL+5×200 mL) and brine (200 mL), dried (Na_2SO_4), filtered, and concentrated to give the title compound (49.0 g) as colorless oil, which was used without further purification in Step B below.

^{1}H NMR (DMSO-d_6): δ 3.28 (s, 3H, OCH_3), 3.53 (d, 2H, $J_{5,4}$=4.5 Hz, H-5a, H-5b), 3.72 (dd, 1H, $J_{3,4}$=3.6 Hz, $J_{3,2}$=6.6 Hz, H-3), 3.99 (ddd, 1H, $J_{2,1}$=4.5 Hz, $J_{2,OH\text{-}2}$=9.6 Hz, H-2), 4.07 (m, 1H, H-4), 4.50 (s, 2H, CH_2Ph), 4.52, 4.60 (2d, 2H, J_{gem}=13.6 Hz, CH_2Ph), 4.54 (d, 1H, OH-2), 4.75 (d, 1H, H-1), 7.32-7.45, 7.52-7.57 (2m, 10H, 2Ph).

^{13}C NMR (DMSO-d_6) δ 55.40, 69.05, 69.74, 71.29, 72.02, 78.41, 81.45, 103.44, 127.83, 127.95, 129.05, 129.28, 131.27, 131.30, 133.22, 133.26, 133.55, 133.67, 135.45, 135.92.

Step B: 3,5-Bis-O-(2,4-dichlorophenylmethyl)-1-O-methyl-α-D-erythro-pentofuranos-2-ulose

To an ice-cold suspension of Dess-Martin periodinane (50.0 g, 118 mmol) in anhydrous CH_2Cl_2 (350 mL) under argon (Ar) was added a solution of the compound from Step A (36.2 g, 75 mmol) in anhydrous CH_2Cl_2 (200 mL) dropwise over 0.5 h. The reaction mixture was stirred at 0° C. for 0.5 h and then at room temperature for 3 days. The mixture was diluted with anhydrous Et_2O (600 mL) and poured into an ice-cold mixture of $Na_2S_2O_3.5H_2O$ (180 g) in saturated aqueous $NaHCO_3$ (1400 mL). The layers were separated, and the organic layer was washed with saturated aqueous $NaHCO_3$ (600 mL), water (800 mL) and brine (600 mL), dried ($MgSO_4$), filtered and evaporated to give the title compound (34.2 g) as a colorless oil, which was used without further purification in Step C below.

^{1}H NMR ($CDCl_3$) δ 3.50 (s, 3H, OCH_3), 3.79 (dd, 1H, $J_{5a,5b}$=11.3 Hz, $J_{5a,4}$=3.5 Hz, H-5a), 3.94 (dd, 1H, $J_{5b,4}$=2.3 Hz, H-5b), 4.20 (dd, 1H, $J_{3,1}$=1.3 Hz, $J_{3,4}$=8.4 Hz, H-3), 4.37 (ddd, 1H, H-4), 4.58, 4.69 (2d, 2H, J_{gem}=13.0 Hz, CH_2Ph), 4.87 (d, 1H, H-1), 4.78, 5.03 (2d, 2H, J_{gem}=12.5 Hz, CH_2Ph), 7.19-7.26, 7.31-7.42 (2m, 10H, 2Ph).

^{13}C NMR (DMSO-d_6) δ 55.72, 69.41, 69.81, 69.98, 77.49, 78.00, 98.54, 127.99, 128.06, 129.33, 129.38, 131.36, 131.72, 133.61, 133.63, 133.85, 133.97, 134.72, 135.32, 208.21.

Step C: 3,5-Bis-O-(2,4-dichlorophenylmethyl)-2-C-methyl-1-O-methyl-α-D-ribofuranose

To a solution of MeMgBr in anhydrous Et_2O (0.48 M, 300 mL) at −55° C. was added dropwise a solution of the compound from Step B (17.40 g, 36.2 mmol) in anhydrous Et_2O (125 mL). The reaction mixture was allowed to warm to −30° C. and stirred for 7 h at −30° C. to −15° C., then poured into ice-cold water (500 mL) and the mixture vigorously stirred at room temperature for 0.5 h. The mixture was filtered through a Celite pad (10×5 cm) which was thoroughly washed with Et_2O. The organic layer was dried ($MgSO_4$), filtered and concentrated. The residue was dissolved in hexanes (~30 mL), applied onto a silica gel column (10×7 cm, prepacked in hexanes) and eluted with hexanes and hexanes/EtOAc (9/1) to give the title compound (16.7 g) as a colorless syrup.

^{1}H NMR (CDCl$_3$): δ 1.36 (d, 3H, $J_{Me,OH}$=0.9 Hz, 2C-Me), 3.33 (q, 1H, OH), 3.41 (d, 1H, $J_{3,4}$=3.3 Hz), 3.46 (s, 3H, OCH$_3$), 3.66 (d, 2H, $J_{5,4}$=3.7 Hz, H-5a, H-5b), 4.18 (apparent q, 1H, H-4), 4.52 (s, 1H, H-1), 4.60 (s, 2H, CH$_2$Ph), 4.63, 4.81 (2d, 2H, J_{gem}=13.2 Hz, CH$_2$Ph), 7.19-7.26, 7.34-7.43 (2m, 10H, 2Ph).

^{13}C NMR (CDCl$_3$): δ 24.88, 55.45, 69.95, 70.24, 70.88, 77.06, 82.18, 83.01, 107.63, 127.32, 129.36, 130.01, 130.32, 133.68, 133.78, 134.13, 134.18, 134.45, 134.58.

Step D: 4-Chloro-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C-methyl-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine

To a solution of the compound from Step C (9.42 g, 19 mmol) in anhydrous dichloromethane (285 mL) at 0° C. was added HBr (5.7 M in acetic acid, 20 mL, 114 mmol) dropwise. The resulting solution was stirred at 0° C. for 1 h and then at rt for 3 h, evaporated in vacuo and co-evaporated with anhydrous toluene (3×40 mL). The oily residue was dissolved in anhydrous acetonitrile (50 mL) and added to a solution of sodium salt of 4-chloro-1H-pyrrolo[2,3-d]pyrimidine [for preparation, see *J. Chem. Soc.,* 131 (1960)] in acetonitrile [generated in situ from 4-chloro-1H-pyrrolo[2,3-d]pyrimidine (8.76 g, 57 mmol) in anhydrous acetonitrile (1000 mL), and NaH (60% in mineral oil, 2.28 g, 57 mmol), after 4 h of vigorous stirring at room temperature]. The combined mixture was stirred at room temperature for 24 h, and then evaporated to dryness. The residue was suspended in water (250 mL) and extracted with EtOAc (2×500 mL). The combined extracts were washed with brine (300 mL), dried over Na$_2$SO$_4$, filtered and evaporated. The crude product was purified on a silica gel column (10 cm×10 cm) using ethyl acetate/hexane (1:3 and 1:2) as the eluent. Fractions containing the product were combined and evaporated in vacuo to give the desired product (5.05 g) as a colorless foam. ^{1}H NMR (CDCl$_3$): δ 0.93 (s, 3H, CH$_3$), 3.09 (s, 1H, OH), 3.78 (dd, 1H, $J_{5',5''}$=10.9 Hz, $J_{5',4}$=2.5 Hz, H-5'), 3.99 (dd, 1H, $J_{5'',4}$=2.2 Hz, H-5"), 4.23-4.34 (m, 2H, H-3', H-4'), 4.63, 4.70 (2d, 2H, J_{gem}=12.7 Hz, CH$_2$Ph), 4.71, 4.80 (2d, 2H, J_{gem}=12.1 Hz, CH$_2$Ph), 6.54 (d, 1H, $J_{5,6}$=3.8 Hz, H-5), 7.23-7.44 (m, 10H, 2Ph).

^{13}C NMR (CDCl$_3$): δ 21.31, 69.10, 70.41, 70.77, 79.56, 80.41, 81.05, 91.11, 100.57, 118.21, 127.04, 127.46, 127.57, 129.73, 129.77, 130.57, 130.99, 133.51, 133.99, 134.33, 134.38, 134.74, 135.21, 151.07, 151.15, 152.47.

Step E: 4-Chloro-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

To a solution of the compound from Step D (5.42 g, 8.8 mmol) in dichloromethane (175 mL) at −78° C. was added boron trichloride (1M in dichloromethane, 88 mL, 88 mmol) dropwise. The mixture was stirred at −78° C. for 2.5 h, then at −30° C. to −20° C. for 3 h. The reaction was quenched by addition of methanol/dichloromethane (1:1) (90 mL) and the resulting mixture stirred at −15° C. for 30 min., then neutralized with aqueous ammonia at 0° C. and stirred at room temperature for 15 min. The solid was filtered and washed with CH$_2$Cl$_2$/MeOH (1/1, 250 mL). The combined filtrate was evaporated, and the residue was purified by flash chromatography over silica gel using CH$_2$Cl$_2$ and CH$_2$Cl$_2$:MeOH (99:1, 98:2, 95:5 and 90:10) gradient as the eluent to furnish desired compound (1.73 g) as a colorless foam, which turned into an amorphous solid after treatment with MeCN.

^{1}H NMR (DMSO-d$_6$) δ 0.64 (s, 3H, CH$_3$), 3.61-3.71 (m, 1H, H-5'), 3.79-3.88 (m, 1H, H-5"), 3.89-4.01 (m, 2H, H-3', H-4'), 5.15-5.23 (m, 3H, 2'-OH, 3'-OH, 5'-OH), 6.24 (s, 1H, H-1'), 6.72 (d, 1H, $J_{5,6}$=3.8 Hz, H-5), 8.13 (d, 1H, H-6), 8.65 (s, 1H, H-2).

^{13}C NMR (DMSO-d$_6$) δ 20.20, 59.95, 72.29, 79.37, 83.16, 91.53, 100.17, 117.63, 128.86, 151.13, 151.19, 151.45.

Step F: 4-Amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

To the compound from Step E (1.54 g, 5.1 mmol) was added methanolic ammonia (saturated at 0° C.; 150 mL). The mixture was heated in a stainless steel autoclave at 85° C. for 14 h, then cooled and evaporated in vacuo. The crude mixture was purified on a silica gel column with CH$_2$Cl$_2$/MeOH (9/1) as eluent to give the title compound as a colorless foam (0.8 g), which separated as an amorphous solid after treatment with MeCN. The amorphous solid was recrystallized from methanol/acetonitrile; m.p. 222° C.

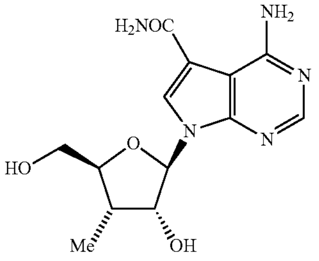
^{1}H NMR (DMSO-d$_6$): δ 0.62 (s, 3H, CH$_3$), 3.57-3.67 (m, 1H, H-5'), 3.75-3.97 (m, 3H, H-5", H-4', H-3'), 5.00 (s, 1H, 2'-OH), 5.04 (d, 1H, $J_{3'OH,3'}$=6.8 Hz, 3'-OH), 5.06 (t, 1H, $J_{5'OH,5',5}$=5.1 Hz, 5'-OH), 6.11 (s, 1H, H-1'), 6.54 (d, 1H, $J_{5,6}$=3.6 Hz, H-5), 6.97 (br s, 2H, NH$_2$), 7.44 (d, 1H, H-6), 8.02 (s, 1H, H-2).

^{13}C NMR (DMSO-d$_6$): δ 20.26, 60.42, 72.72, 79.30, 82.75, 91.20, 100.13, 103.08, 121.96, 150.37, 152.33, 158.15.

LC-MS: Found: 279.10 (M−H$^+$); calc. for $C_{12}H_{16}N_4O_4$+H$^+$: 279.11.

EXAMPLE 63

4-Amino-7-(3-deoxy-3-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide



Step A: 4-Amino-6-bromo-7-(2-O-acetyl-5-O-benzoyl-3-deoxy-3-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile

BSA (0.29 mL, 2.0 mmol) was added into a stirred suspension of 4-amino-6-bromo-5-cyano-1H-pyrrolo[2,3-d]pyrimidine (0.24 g, 1 mmol; prepared according to *Nucleic Acid Chemistry*, Part N, Townsend, L. B. and Tipson, R. S.; Ed.; Wiley-Interscience: New York, 1991, pp. 16-17 and *Synthetic Commun.* 1998, 28, 3835) in dry acetonitrile (10 mL) at room temperature under argon. After 15 min, 1,2-di-O-acetyl-5-O-benzoyl-3-deoxy-3-methyl-D-ribofuranose (J. Med. Chem. (1976), 19, 1265) (0.36 g, 1.0 mmol) was added along with TMSOTf (0.54 g, 3 mmol). The mixture was stirred at room temperature for 5 min and then at 80° C. for 0.5 h. The solution was cooled, diluted with ethyl acetate (50 mL) and poured into ice-cold saturated aqueous NaHCO$_3$ (15 mL).

The layers were separated. The organic layer was washed with brine (15 mL), dried (Na_2SO_4) and then evaporated. The residue was purified on silica gel column using a solvent system of hexanes/EtOAc: 3/1. Appropriate fractions were collected and evaporated to provide the title compound as colorless foam (0.21 g).

Step B: 4-Amino-7-(2-O-acetyl-5-O-benzoyl-3-deoxy-3-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carbonitrile

To a suspension of the title compound from Step A (183 mg, 0.35 mmol) in EtOH (9 mL) were added ammonium formate (0.23 g, 3.6 mmol) and 10% palladium on activated carbon (20 mg) and the mixture was heated at reflux for 1.5 h. The hot reaction mixture was filtered through Celite and washed with hot EtOH. The solvent was removed and the residue treated with MeOH. The pale yellow solid was filtered thus yielding 105 mg of pure title compound. The filtrate was evaporated and purified on a silica gel column with a solvent system of CH_2Cl_2/MeOH: 50/1 to afford an additional 63 mg of title compound as a white solid.

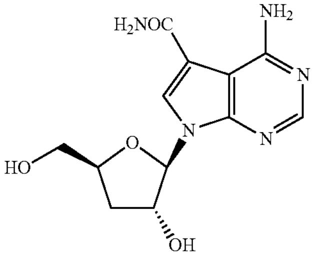
Step C: 4-Amino-7-(3-deoxy-3-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-5-carboxamide

A mixture of the compound from Step B (51 mg, 0.12 mmol), ethanolic ammonia (5 mL, saturated at 0° C.), aqueous ammonia (5 mL, 30%) and aqueous hydrogen peroxide (1 mL, 35%) was stirred room temperature for 8 h. The solution was evaporated and the residue purified on silica gel column with a solvent system of CH_2Cl_2/MeOH: 10/1 to give the title compound as a white solid (28 mg).

^{1}H-MNR (CD_3OD): δ 1.12 (d, 3H, J=6.8 Hz), 2.40 (m, 1H), 3.76 (dd, 1H, J_1=12.8 Hz, J_2=4.0 Hz), 3.94-4.04 (m, 2H), 4.33 (d, 1H, J=5.4 Hz), 6.13 (s, 1H), 8.11 (s, 1H), 8.16 (s, 1H).

EXAMPLE 64

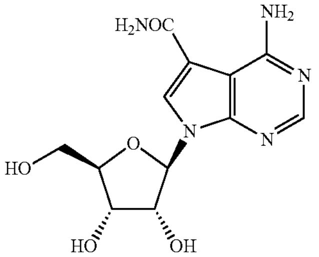
4-Amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carboxamide



This compound was prepared following the procedures described in *J. Med. Chem.* 26: 25 (1983).

EXAMPLE 65

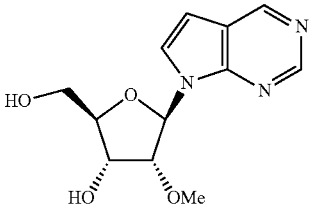
4-Amino-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carboxamide (Sangivamycin)



This compound was obtained from commercial sources.

EXAMPLE 66

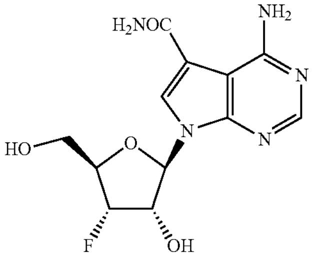
7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



This compound was prepared following the procedures described in *J. Org. Chem.* 39: 1891 (1974).

EXAMPLE 67

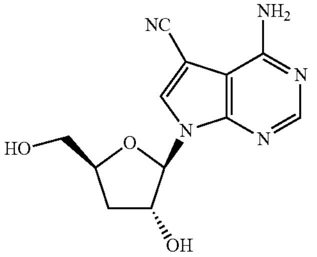
4-Amino-7-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carboxamide



This compound was prepared following the procedures described in *Chem. Pharm. Bull.* 41: 775 (1993).

EXAMPLE 68

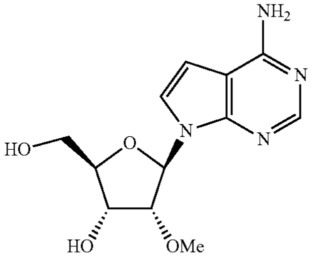
4-Amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile



This compound was prepared following the procedures described in *J. Med. Chem.* 30: 481 (1987).

EXAMPLE 69

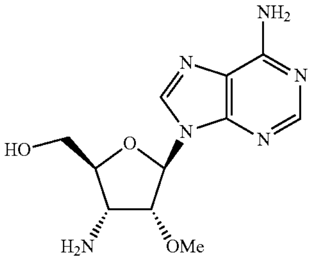
4-Amino-7-(2-O-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



This compound was prepared following the procedures described in *J. Org. Chem.* 39: 1891 (1974).

EXAMPLE 70

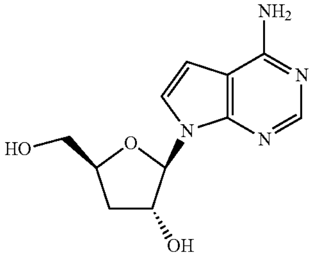
3'-Amino-3'-deoxy-2'-O-methyladenosine



This compound is obtained by the methylation of appropriately protected 3'-amino-3'-deoxyadenosine derivative (Example 54).

EXAMPLE 71

4-Amino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



This compound was prepared following the following procedure described in *Can. J. Chem.* 55: 1251 (1977).

EXAMPLE 72

General Process to SATE Prodrug Moiety

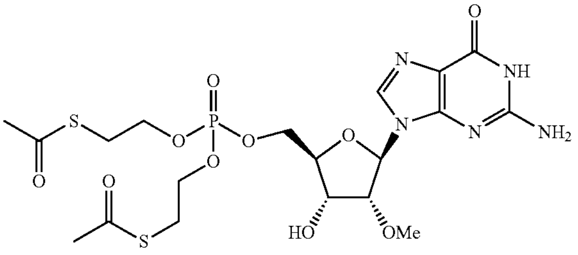
S-Acyl-2-Thioethyl (SATE) pronucleotides are discussed in C. R. Wagner, V. V. Iyer, and E. J. McIntee, "Pronucleotides: Toward the In Vivo Delivery of Antiviral and Anticancer Nucleotides," *Med. Res. Rev.*, 20:1-35 (2000), which is incorporated by reference herein in its entirety. SATE derivatives of nucleosides are also disclosed U.S. Pat. Nos. 5,770,725; 5,849,905; and 6,020,482, the contents of each of which are incorporated by reference herein in their entirety.

Bis(S-acetyl-2-thioethyl)-N,N-diisopropylphosphoramidite

2-Mercaptoethanol (5 g, 64 mmol) was dissolved in CH_2Cl_2 (50 mL). To this solution was added triethylamine (7.67 mL, 57.6 mmol), and the reaction mixture was cooled in an ice bath to 0° C. Acetic anhydride (4.54 mL, 48 mmol) was added dropwise in 10 min, and the reaction mixture was stirred for 1 h at 0° C. The reaction mixture was then allowed to come to room temperature over a period of 2 h. The reaction mixture was diluted with CH_2Cl_2 (50 mL), washed with water (75 mL), 5% aqueous $NaHCO_3$ (75 mL) and brine (75 mL). The organic phase was dried over anhydrous Na_2SO_4 and concentrated in vacuo to give an oil. The oil was then dissolved in anhydrous THF (40 mL) and anhydrous triethylamine (7.76 mL) was added. To this mixture was added activated molecular sieves (4 Å) and was kept at room temperature for 10 min. The reaction mixture was cooled in an ice bath to 0° C. and diisopropylphosphoramidous dichloride (6.47 g, 32.03 mmol) was added. The reaction mixture was stirred at 0° C. for 2 h under inert atmosphere. Hexane (40 mL) was added to the reaction mixture and the precipitate formed was filtered. The filtrate was concentrated to one fourth of the volume, purified by loaded silica gel column chromatography and eluted with hexane containing 3% triethylamine and incremental amount of ethyl acetate (0 to 7%) to give the title compound as an oil (2.36 g). ^1H NMR ($CDCl_3$): δ 1.17 (s, 6H), 1.21 (s, 6H), 2.36 (s, 6H), 3.14 (t, J=6.44 Hz), 3.51-3.84 (m, 6H); ^{13}C NMR ($CDCl_3$): δ 24.47, 24.61, 30.48, 42.85, 43.1, 61.88, 62.23, 195.26; ^{13}P NMR ($CDCl_3$): δ 146.96.

EXAMPLE 73

2'-O-Methylguanosine-5'-[bis-(S-acetyl-2-thioethyl) phosphate]



Step A: N^2-(4-monomethoxytrityl)-2'-O-methylguanosine-5'-[bis-(S-acetyl-2-thioethyl)phosphate]

N^2-(4-monomethoxytrityl)-2'-O-methylguanosine (0.74 g, 1.31 mmol) was mixed with 1H-tetrazole (0.061 g, 0.87 mmol) and dried over P_2O_5 in vacuo overnight. To this mixture was added anhydrous acetonitrile (8 mL). To the turbid solution, bis(S-acetyl-2-thioethyl)N,N-diisopropylphosphoramidite (0.3 g, 0.87 mmol) was added slowly and the reaction mixture was stirred at ambient temperature under inert atmosphere for 2 h. Solvent was removed in vacuo. The residue was cooled to −40° C. and a solution of 3-chloroperbenzoic acid (0.2 g) in CH_2Cl_2 (7 mL) was added. The solution was allowed to warm up to room temperature over 1 h. Sodium hydrogensulfite (10% aqueous solution, 2 mL) was added to reduce the excess of 3-chloroperbenzoic acid. The organic phase separated, diluted with CH_2Cl_2 (20 mL), washed with saturated aqueous Na_2CO_3 (10 mL), water (10 mL), dried over Na_2SO_4 and evaporated to dryness. The residue was purified by silica gel column chromatography and eluted with CH_2Cl_2 containing incremental amount of MeOH (5 to 10%) as eluent to yield the title compound (0.36 g) as a foam.

^{1}H NMR (DMSO-d_6): δ 2.35 (s, 6H), 2.97 (s, 3H), 3.11 (t, 4H, J=6.0 Hz), 3.5 (m, 1H), 3.74 (s, 3H), 3.72-3.83 (m, 2H), 3.97-4.11 (m, 6H), 5.1 (d, 1H, J=6.4 Hz), 5.29 (d, 1H, J=3.1 Hz), 6.89 (d, 2H, J=8.8 Hz), 7.15-7.37 (m, 12H), 7.68 (s, 1H), 7.73 (s, 1H), 10.72 (s, 1H); ^{13}C NMR ($CDCl_3$): δ 30.36, 55.38, 57.99, 66.08, 66.19, 67.22, 69.15, 70.49, 81.18, 81.57, 86.64, 113.04, 117.99, 126.66, 127.71, 128.67, 130.04, 136.09, 136.56, 144.51, 144.82, 149.52, 151.29, 158.15, 194.56; ^{13}P NMR ($CDCl_3$): δ −2.04; MS (API-ES) 852.10 $[M-H]^+$.

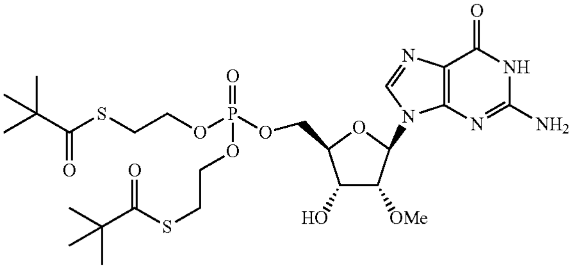
Step B: 2'-O-methylguanosine-5'-[bis-(S-acetyl-2-thioethyl)phosphate]

N^2-(4-monomethoxytrityl)-2'-O-methylguanosine-5'-[bis-(S-acetyl-2-thioethyl)phosphate] (0.2 g, 0.23 mmol) was dissolved in acetic acid:MeOH:H_2O, 3:6:1 and heated at 55° C. for 24 h. Solvent was removed and the residue was purified by HPLC on reverse phase column (Hamilton PRP-1, 250×22 mm, A=Acetonitrile, B=H_2O 20 to 100 B in 65 min, flow 10 mL min^{-1}). Fractions containing the product were pooled together and evaporated to give the title compound (40% yield).

^{13}P NMR ($CDCl_3$): δ −0.72; MS (API-ES) m/z 582.1 $[M+H]^+$.

EXAMPLE 74

2'-O-Methylguanosine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate]



Step A: Bis(S-pivaloyl-2-thioethyl)-N,N-diisopropylphosphoramidite

S-pivaloyl-2-thioethanol (6.3 g, 39.6 mmol) was dissolved in anhydrous THF (100 mL). To this solution was added activated molecular sieves (4A°) and kept at room temperature for 30 min. Anhydrous triethylamine (7.9 mL, 59.4 mmol) was added and the reaction mixture was cooled in an ice bath to 0° C. To this mixture diisopropylphosphoramidous dichloride (4 g, 19.8 mmol) was added dropwise. The mixture was stirred the reaction mixture at 0° C. for 2 h under inert gas atmosphere. Hexane (100 mL) was added to the reaction mixture, and the precipitate formed was filtered. The filtrate was concentrated to one fourth of the volume. This was purified by flash silica gel column chromatography using hexane containing 2% triethylamine and incremental amount of ethyl acetate (0 to 3%) as eluent to give the title compound as an oil (5.23 g).

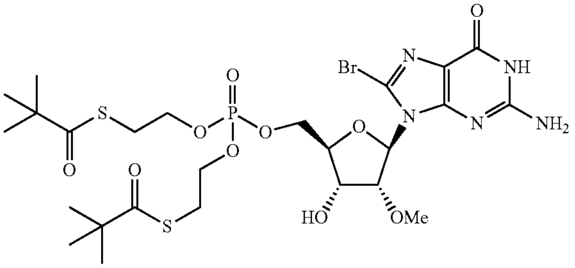
^{1}H NMR ($CDCl_3$): δ 1.13-1.31 (m, 30H), 1.21 (s, 6H), 3.09 (t, J=6.6 Hz, 4H), 3.51-3.84 (m, 6H); ^{13}C NMR ($CDCl_3$): δ 24.47, 24.61, 27.32, 30.00, 42.85, 43.1, 46.32, 61.98, 62.33, 206.1; ^{13}P NMR ($CDCl_3$): δ 148.51.

Step B: 2'-O-methylguanosine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate]

N^2-(4-monomethoxytrityl)-2'-O-methylguanosine (0.6 g, 1.05 mmol) was mixed with 1H-tetrazole (0.05 g, 0.7 mmol) and dried over P_2O_5 in vacuo overnight. To this mixture anhydrous acetonitrile (13.8 mL) was added. The reaction mixture was cooled to 0° C. in an ice bath and bis(S-pivaloyl-2-thioethyl)N,N-diisopropylphosphoramidite (0.32 g, 0.7 mmol) was added slowly. The reaction mixture was stirred at 0° C. for 5 minutes. The ice bath was removed and the reaction mixture was allowed to stir at room temperature under an inert atmosphere for 2 h. Solvent was removed in vacuo. The residue was cooled to −40° C. and a solution of 3-chloroperbenzoic acid (0.24 g, 1.4 mmol, 57-80%) in CH_2Cl_2(10 mL) was added. The solution was allowed to warm up to −10° C. over 1 h. Sodium hydrogensulfite (10% aqueous solution, 10 mL) was added to reduce the excess of 3-chloroperbenzoic acid. The organic phase separated, diluted with CH_2Cl_2 (50 mL), washed with saturated aqueous Na_2CO_3 (40 mL), water (40 mL), dried over Na_2SO_4 and evaporated to dryness. The residue was chromatographed on a flash silica gel column using a CH_2Cl_2 containing incremental amount of MeOH (0 to 5%) as eluent. Fractions containing the product were pooled together and evaporated. The residue was dissolved in a solution of acetic acid/water/methanol (10 mL, 3:1:6) and heated at 55° C. for 24 h. Evaporated the solution in vacuum to get an oil. The oil was dissolved in 20% MeOH in water and purified by HPLC on C-18 column (Luna C-18, 250×2.12 mm, A=water, B=acetonitrile, 20 to 10% B in 65 min., flow 10 mL min^{-1}, λ 260 nm) to yield the title compound (0.082 g). ^1H NMR (DMSO-d_6): δ 1.18 (s, 18H), 3.08 (m, 4H), 3.33 (s, 3H) 3.94-4.10 (m, 6H), 4.14-4.21 (m, 2H), 4.29 (m, 1H), 5.42 (d, 1H, J=5.4 Hz), 5.81 (d, 1H, J=5.8 Hz), 6.49 (bs, 2H), 7.86 (s, 1H), 10.66 (bs, 1H); ^{13}P NMR (DMSO-d_6): δ −0.71; MS (API-ES) m/z 664.2 $[M-H]^-$.

EXAMPLE 75

8-Bromo-2'-O-methylguanosine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate]

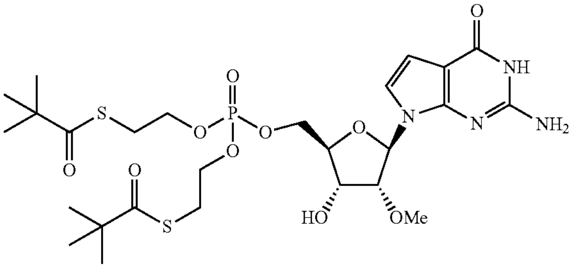


This compound was synthesized according to the procedure used for the synthesis of Example 74 starting with 8-bromo-N^2-(4-monomethoxytrityl)-2'-O-methylguanosine (0.46 g, 0.63 mmol). Other reagents used were 1H-tetrazole (0.034 g, 0.49 mmol), bis(S-pivaloyl-2-thioethyl)N,N-diisopropylphosphoramidite (0.22 g, 0.49 mmol), acetonitrile (8.3 ml), 3-chloroperbenzoic acid (0.17 g, 0.98 mmol, 57-80%) in CH_2Cl_2 (4 mL). The title compound was isolated in 13% yield (0.061 g).

^1H NMR (DMSO-d_6): δ 1.14 and 1.16 (m, 18H), 3.06 (m, 4H), 3.32 (s, 3H) 3.96-4.06 (m, 5H), 4.18-4.3 (m, 2H), 4.46 (d, 1H, J=2.4 Hz), 4.66 (t, 1H, J=2.6 Hz), 5.37 (d, 1H, J=2.6 Hz), 5.78 (d, 1H, J=2.8 Hz), 6.62 (bs, 2H), 10.99 (bs, 1H); ^{13}P NMR (DMSO-d_6) 8-0.79; MS (API-ES) m/z 742.13 and 744.13 $[M-H]^-$.

EXAMPLE 76

2-Amino-3,4-dihydro-7-(2-O-methyl-β-D-ribofuranosyl)-4-oxo-7H-pyrrolo[2,3-d]pyrimidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate]

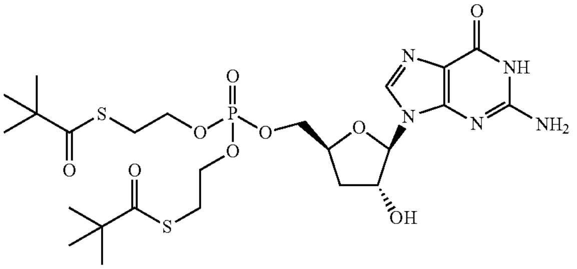


This compound was synthesized according to the procedure used for the synthesis of Example 74 starting with 7-deaza-N^2-(4-monomethoxytrityl)-2'-O-methylguanosine (0.47 g, 0.82 mmol). Other reagents used were 1H-tetrazole (0.044 g, 0.63 mmol), bis(S-pivaloyl-2-thioethyl) N,N-diisopropylphosphoramidite (0.29 g, 0.63 mmol), acetonitrile (11 mL), 3-chloroperbenzoic acid (0.21 g, 1.26 mmol, 57-80%) in CH_2Cl_2 (5.2 mL). The title compound was isolated in 29% yield (0.158 g).

^1H NMR (DMSO-d_6): δ 1.14 (s, 18H), 3.06 (m, 4H), 3.31 (s, 3H) 3.96-4.26 (m, 9H), 5.35 (d, 1H, J=2.6 Hz), 5.78 (d, 1H, J=5.2 Hz), 5.99 (d, 1H, J=6.6 Hz), 6.27 (m, 3H), 6.86 (d, 1H, J=3.6 Hz), 10.39 (s, 1H); ^{13}P NMR (DMSO-d_o): 8-0.72; MS (API-ES) m/z 663.20 $[M-H]^-$; HRMS Calcd for $C_{26}H_{42}N_4O_{10}PS_2$ 665.2074 found 665.2071.

EXAMPLE 77

3'-Deoxyguanosine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate]

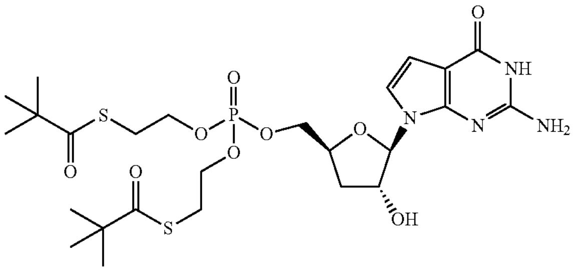


N^2-(4-Monomethoxytrityl)-3'-deoxyguanosine (0.20 g, 0.35 mmol) was mixed with 1H-tetrazole (0.019 g, 0.27 mmol) and dried over P_2O_5 in vacuo overnight. To this mixture anhydrous acetonitrile (4.7 mL) was added to give a turbid solution. The reaction mixture was cooled to 0° C. in an ice bath and bis(S-pivaloyl-2-thioethyl)N,N-diisopropylphosphoramidite (0.12 g, 0.27 mmol) was added slowly. The reaction mixture was stirred at 0° C. for 5 minutes. The ice bath was removed and the reaction mixture was allowed to come to room temperature. The reaction mixture was stirred at room temperature under an inert gas atmosphere for 2 h. Solvent was removed in vacuo. The residue was cooled to −40° C. and a solution of 3-chloroperbenzoic acid (0.12 g, 0.7 mmol, 57-80%) in CH_2Cl_2 (2.2 mL) was added. The solution was allowed to warm up to −10° C. over 1 h. Sodium hydrogensulfite (10% aqueous solution, 2 mL) was added to reduce the excess of 3-chloroperbenzoic acid. The organic phase was separated, diluted with CH_2Cl_2 (30 mL), washed with saturated aqueous Na_2CO_3 (20 mL), water (20 mL), dried over Na_2SO_4 and evaporated to dryness. The residue was chromatographed on a flash silica gel column using CH_2Cl_2 containing incremental amount of MeOH (0 to 5%) as eluent. Fractions containing the product were pooled and evaporated. The residue was dissolved in a solution of acetic acid/water/methanol (5 mL, 3:1:6) and heated at 55° C. for 24 h. Evaporated the solution in vacuum to get an oil. The oil was dissolved in 20% MeOH in water and purified by HPLC on C-18 column (Luna C-18, 250×2.12 mm, A=water, B=acetonitrile, 20 to 10% B in 65 min., flow 10 mL min^{-1}, λ 260 nm) to yield the title compound (0.027 g).

^1H NMR (DMSO-d_6): δ 1.15 (s, 18H), 1.92-2.01 (m, 1H), 2.17-2.28 (m, 1H), 3.04 (t, 4H, J=6.2 Hz), 3.91-4.23 (m, 6H), 4.37-4.55 (m, 2H), 5.67 (m, 2H), 6.45 (bs, 2H), 7.75 (s, 1H), 10.61 (s, 1H); ^{13}P NMR (DMSO-d_6): δ −0.75; MS (API-ES) m/z 634.2 $[M-H]^-$.

EXAMPLE 78

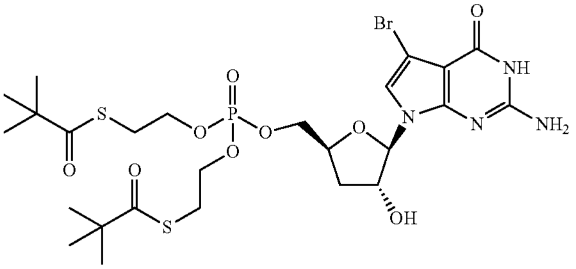
2-Amino-7-(3-deoxy-β-D-ribofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo[2,3-d]pyrimidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate]



2-(4-Monomethoxytritypamino-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one (0.30 g, 0.52 mmol) was mixed with 1H-tetrazole (0.028 g, 0.40 mmol) and dried over P_2O_5 in vacuo overnight. To this mixture anhydrous acetonitrile (7 mL) was added, and the solution was cooled to 0° C. in an ice bath. Bis(S-pivaloyl-2-thioethyl)-N,N-diisopropylphosphoramidite (0.18 g, 0.40 mmol) was added slowly. The reaction mixture was allowed to come to at room temperature and stirred at room temperature under an inert atmosphere for 2 h. The solvent was removed in vacuo. The residue was cooled to −40° C., and a solution of 3-chloroperbenzoic acid (0.14 g, 0.8 mmol, 57-80%) in CH_2Cl_2 (5 mL) was added. The solution was allowed to warm up to −10° C. over 2 h. Sodium hydrogensulfite (10% aqueous solution, 5 mL) was added to reduce the excess of 3-chloroperbenzoic acid. The organic phase was separated, diluted with CH_2Cl_2 (50 mL), washed with saturated aqueous Na_2CO_3 (40 mL), water (40 mL), dried over Na_2SO_4 and evaporated to dryness. The residue was chromatographed on a flash silica gel column using CH_2Cl_2 containing incremental amount of MeOH (0 to 5%) as eluent. Fractions containing the product were pooled and evaporated. The residue was dissolved in a solution of acetic acid/water/methanol (10 mL, 3:1:6) and heated at 55° C. for 24 h. The solution was evaporated to give an oil. The oil was dissolved in 20% MeOH in water and purified by HPLC on C-18 column (Luna C18,250×2.12 mm, A=water, B=acetonitrile 20 to 10% B in 65 mL, flow 10 mL/min, λ 260 nm) to give the title compound (0.053 g). ^1H NMR (DMSO-d_6): δ 1.16 (s, 18H), 1.91-2.01 (m, 1H), 2.17-2.25 (m, 1H), 3.05 (t, 4H, J=6.2 Hz), 3.92-4.2 (m, 6H), 4.35 (bs, 2H), 5.56 (d, 1H, J=4.2 Hz), 5.86 (d, 1H, J=2.4 Hz), 6.24 (m, 3H), 6.77 (d, 1H, J=3.6 Hz), 10.36 (s, 1H); ^{13}P NMR (DMSO-d_6): δ −0.89; HRMS (MALDI) Calcd for $C_{25}H_{39}N_4O_9PS_2$.635.1969 found 635.1964.

EXAMPLE 79

2-Amino-5-bromo-7-(3-deoxy-β-D-ribofuranosyl)-3,4-dihydro-4-oxo-7H-pyrrolo[2,3-d]pyrimidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate]

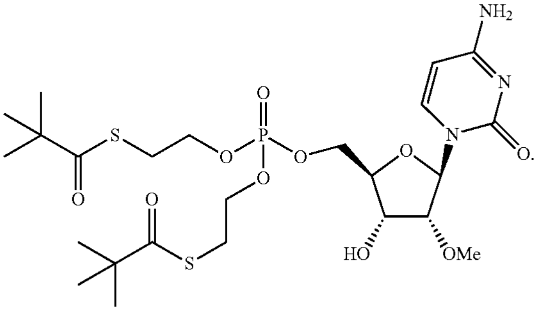


2-(4-Monomethoxytritypamino-5-bromo-7-(3-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one (0.066 g, 0.17 mmol) was mixed with imidazole triflate (0.017 g, 0.17 mmol) and dried over P_2O_5 in vacuo overnight. To this mixture anhydrous acetonitrile (7 mL) and bis(S-pivaloyl-2-thioethyl)N,N-diisopropylphosphoramidite (0.97 g, 0.24 mmol) were added slowly. The reaction mixture was stirred under an inert atmosphere for 18 h. Solvent was removed in vacuo. The residue was cooled to −40° C. and a solution of 3-chloroperbenzoic acid (0.059 g, 0.34 mmol, 57-80%) in CH_2Cl_2 (2 mL) was added. The solution was allowed to warm up to −10° C. over 2 h. Sodium hydrogensulfite (10% aqueous solution, 5 mL) was added to reduce the excess of 3-chloroperbenzoic acid. The organic phase was separated, diluted with CH_2Cl_2 (30 mL), washed with saturated aqueous Na_2CO_3 (20 mL), water (20 mL), dried over Na_2SO_4 and evaporated to dryness. The residue was chromatographed on flash silica gel column using CH_2Cl_2 containing incremental amount of MeOH (0 to 5%) as eluent. Fractions containing the product were pooled and evaporated. The residue was dissolved in a solution of acetic acid/water/methanol (3 mL, 3:1:6) and heated at 55° C. for 24 h. The solution was evaporated to give an oil. The oil was dissolved in 20% MeOH in water and purified by HPLC on C-18 column (Luna C18,250×2.12 mm, A=water, B=acetonitrile 20 to 10% B in 65 mL, flow 10 mL min^{-1}, λ 260 nm) to afford the title compound (0.036 g).

^{1}H NMR (DMSO-d$_6$): δ 1.17 (s, 18H), 1.87-2.03 (m, 1H), 2.17-2.26 (m, 1H), 3.05 (t, 4H, J=6.4 Hz), 3.92-4.2 (m, 6H), 4.37 (bs, 2H), 5.70 (d, 1H, J=4.4 Hz), 5.85 (d, 1H, J=2.6 Hz), 6.36 (bs, 2H), 6.93 (s, 1H), 10.51 (s, 1H); ^{13}P NMR (DMSO-d$_6$): δ −0.89; MS (AP-ES) m/z 711.11 and 713.09 [M−H]$^-$; HRMS (MALDI) Calcd for $C_{25}H_{38}BrN_4O_9PS_2$.713.1074 and 715.1074 found 713.1081 and 715.102.

EXAMPLE 80

2'-O-Methylcytidine-5'-[bis-(S-pivaloyl-2-thioethyl) phosphate]

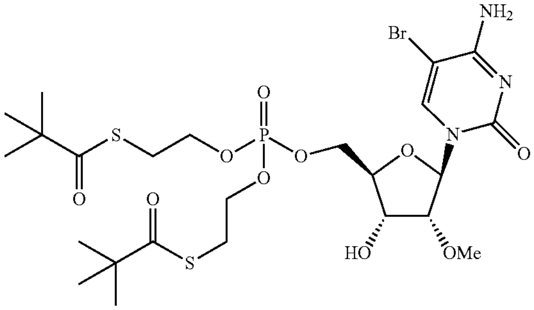


N^4-(4,4'-Dimethoxytrityl)-2'-O-methylcytidine (0.49 g, 0.86 mmol) was mixed with 1H-tetrazole (0.06 g, 0.86 mmol) and dried over P_2O_5 in vacuo overnight. To this mixture anhydrous acetonitrile (6 mL) and bis-(S-pivaloyl-2-thioethyl)-N,N-diisopropylphosphoramidite (0.39 g, 0.86 mmol) were added at 0° C. The reaction mixture was allowed to come to room temperature and stirred under an inert atmosphere for 18 h. Solvent was removed in vacuo. The residue was cooled to −40° C. and a solution of 3-chloroperbenzoic acid (0.3 g, 1.72 mmol, 57-80%) in CH_2Cl_2 (5.5 mL) was added. The solution was allowed to warm up to −10° C. over 2 h. Sodium hydrogensulfite (10% aqueous solution, 5 mL) was added to reduce the excess of 3-chloroperbenzoic acid. The organic phase was separated, diluted with CH_2Cl_2 (30 mL), washed with saturated aqueous Na_2CO_3 (20 mL), water (20 mL), dried over Na_2SO_4 and evaporated to dryness. The residue was chromatographed on a flash silica gel column using CH_2Cl_2 containing incremental amount of MeOH (0 to 10%) as eluent. Fractions containing the product were pooled and evaporated. The residue was dissolved in a solution of acetic acid/water/methanol (10 mL, 3:1:6) and heated at 55° C. for 24 h. The solution was evaporated to give an oil. The oil was dissolved in 20% MeOH in water and purified by HPLC on C-18 column (Luna C18,250×2.12 mm, A=water, B=acetonitrile 20 to 10% B in 65 mL, flow 10 mL min^{-1}, λ 260 nm) to yield the title compound (0.076 g).

^{1}H NMR (DMSO-d$_6$): δ 1.18 (s, 18H), 3.12 (t, 4H, J=6.4 Hz), 3.39 (s, 3H), 3.69 (t, 1H, J=4.2 Hz), 3.93-4.3 (m, 8H), 5.29 (d, 1H, J=6.2 Hz), 5.72 (d, 1H, J=7.4 Hz), 5.86 (d, 1H, J=4 Hz), 7.21 (bs, 2H), 7.58 (d, 1H, J=7.4 Hz); ^{13}P NMR (CD_3CN): δ −0.64; MS (AP-ES) m/z 625.69 [M+H]$^+$; HRMS (MALDI) Calcd for $C_{24}H_{40}N_3O_{10}PS_2Na$ 648.1785 found 648.1804.

EXAMPLE 81

5-Bromo-2'-O-methylcytidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate]



Step A: 5-Bromo-3'-O-(t-butyldimethyl)silyl-2'-O-methylcytidine

2'-O-Methylcytidine (1.5 g, 5.83 mmol) was mixed with imidazole (3.97 g, 58.32 mmol) and dried in vacuo. This mixture was dissolved in anhydrous DMF (4 mL) and t-butyldimethylsilyl chloride (4.41 g, 29.25 mmol) was added and the reaction mixture was stirred for 18 h at room temperature under an inert atmosphere. Reaction mixture was diluted with water (100 mL) and extracted with ethyl acetate (2×60 mL). The organic phase was dried over anhydrous Na_2SO_4 and evaporated. The residue was purified by silica gel column chromatography and eluted with ethyl acetate/hexane, 6:4. Fractions containing the product were pooled and evaporated. The product obtained (2.76 g) was dissolved in acetonitrile (19.43 mL), LiBr (0.623 g, 7.18 mmol) and stirred to get a clear solution. To this ammonium ceric (IV) nitrate (6.24 g, 11.37 mmol) was added and the reaction mixture was allowed to stir at room temperature for 3 h. Solvent was removed in vacuum. The residue obtained was taken in ethyl acetate (100 mL) and washed with water (80 mL). The organic phase was separated, dried over anhydrous Na_2SO_4 and evaporated. Residue purified by silca gel column chromatography and eluted with 5% MeOH in CH_2Cl_2. The product obtained (2.66 g) was dissolved in 80% acetic acid in water and heated at 50° C. for 6 h. The solvent was removed and the residue purified on a silica gel column and eluted with 5% MeOH in CH_2Cl_2 to give the title compound (0.85 g). ^{1}H NMR (DMSO-d$_6$): δ 0.78 (s, 6H), 0.85 (s, 9H), 3.31 (s, 3H), 3.44-3.6 (m, 2H), 3.69-3.9 (m, 2H), 4.24 (m, 1H), 5.29 (t, 1H, J=4.4 Hz), 5.76 (d, 1H, J=3.2 Hz), 7.06 (bs, 1H), 7.88 (bs, 1H), 8.39 (s, 1H).

Step B: 5-Bromo-2'-O-methylcytidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate]

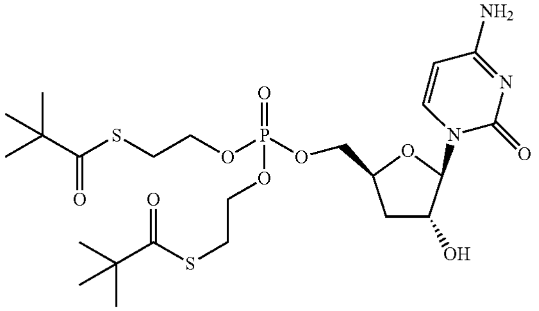
5-Bromo-3'-O-(t-butyldimethyl)silyl-2'-O-methylcytidine (0.093 g, 0.21 mmol) was mixed with 1H-tetrazole (0.03 g, 0.42 mmol) and dried over P_2O_5 in vacuo overnight. To this mixture anhydrous acetonitrile (2 mL). Bis-(S-pivaloyl-2-thioethyl)-N,N-diisopropylphosphoramidite (0.2 g, 0.42 mmol) was added at 0° C. The reaction mixture was allowed to come to room temperature and stirred under an inert atmosphere for 4 h. Solvent was removed in vacuo. The residue was cooled to −40° C. and a solution of 3-chloroperbenzoic acid (0.072 g, 0.42 mmol, 57-80%) in CH_2Cl_2 (2 mL) was added. The solution was allowed to warm up to −10° C. over

2 h. Sodium hydrogensulfite (10% aqueous solution, 2 mL) was added to reduce the excess of 3-chloroperbenzoic acid. The organic phase separated, diluted with CH_2Cl_2 (30 mL), washed with saturated aqueous Na_2CO_3 (20 mL), water (20 mL), dried over Na_2SO_4 and evaporated to dryness. The residue was dissolved in THF (2.1 mL) and triethylamine trihydrofluoride (0.17 g, 1.1 mmol). The reaction mixture was stirred at room temperature for 18 h. The solution was evaporated to give an oil. The oil was dissolved in ethyl acetate (30 mL) and washed with water (20 mL), 5% aqueous $NaHCO_3$ and brine (20 mL). The organic phase was dried over anhydrous Na_2SO_4 and evaporated. The residue was dissolved in 20% MeOH in water and purified by HPLC on C-18 column (Luna C18, 250×2.12 mm, A=water, B=acetonitrile 20 to 10% B in 65 mL, flow 10 mL min^{-1}, λ 260 nm) to give the title compound (0.054 g).

^{1}H NMR (DMSO-d_6): δ 1.17 (s, 18H), 3.11 (t, 4H, J=6.2 Hz), 3.39 (s, 3H), 3.75 (t, 1H, J=4.8 Hz), 3.93-4.3 (m, 8H), 5.23 (d, 1H, J=6.4 Hz), 5.8 (d, 1H, J=3.8 Hz), 7.07 (bs, 1H), 7.89 (s, 1H) 7.94 (bs, 1H); ^{13}P NMR (CD_3CN): δ −0.34; MS (AP-ES) m/z 702.00 and 704.00 $[M-H]^-$; HRMS (MALDI) Calcd for $C_{24}H_{39}BrN_3O_{10}PS_2Na$ 726.0890 and 728.0890 found 726.0893 and 728.086.

EXAMPLE 82

3'-Deoxycytidine-5'-[bis-(S-pivaloyl-2-thioethyl) phosphate]



Step A: N^4-(4,4'-dimethoxytrityl)-3'-deoxycytidine

3'-Deoxycytidine (0.8 g, 3.54 mmol) was mixed with imidazole (2.41 g, 35.4 mmol) and dried over P_2O_5 in vacuum overnight at 40° C. The mixture was dissolved in anhydrous DMF and t-butyldimethylsilyl chloride (2.68 g, 17.78 mmol) was added and the reaction mixture was stirred under an argon atmosphere for 18 h at room temperature. The reaction mixture was diluted with water (100 mL) and extracted with ethyl acetate (2×75 mL). The organic phase was separated, dried over anhydrous Na_2SO_4 and evaporated. The residue was purified by silica gel column chromatography and eluted with ethyl acetate/hexane (6:4) to yield 2',5'-bis(t-butyldimethylsilyl)-3'-deoxycytidine (1.27 g). This was then mixed with DMAP (0.34 g, 2.79 mmol) and dried in vacuum. This mixture was dissolved in anhydrous pyridine (8 mL) and 4,4'-dimethoxytrityl chloride (1.89 g, 5.58 mmol) was added. The reaction mixture was stirred at room temperature under an argon atmosphere for 18 h. Solvent was removed in vacuo. The residue obtained was taken in ethyl acetate (100 mL) and washed with 5% $NaHCO_3$ in water (75 mL) and brine (75 ml). The organic phase was dried over anhydrous Na_2SO_4 and evaporated. The residue obtained was dissolved in THF (28 mL). To this triethylamine trihydrofluoride (2.26 mL, 13.74 mmol) and triethylamine (0.95 mL, 6.87 mmol) were added and stirred at room temperature for 18 h. Solvent was removed and the residue dissolved in ethyl acetate (50 mL), washed with water (50 mL) and 5% $NaHCO_3$ in water (50 mL). The organic phase was dried over anhydrous Na_2SO_4 and evaporated. The residue obtained was purified by silica gel column chromatography and eluted with 5% MeOH in CH_2Cl_2 to yield the title compound (0.66 g).

^{1}H NMR (DMSO-d_6): δ 1.66 (m, 1H), 1.85 (m, 1H), 3.47 (m, 1H), 3.63 (m, 1H), 3.71 (s, 6H), 4.00 (bs, 1H), 4.19 (m, 1H), 4.96 (t, 1H, J=5.2 Hz), 5.39 (bs, 1H), 5.53 (s, 1H), 6.17 (bs, 1H), 6.83 (d, 4H, J=8.8 Hz), 7.04-7.22 (m, 9H), 7.77 (d, 1H, J=7.6 Hz), 8.27 (bs, 1H); MS (AP-ES) m/z 528.1 $[M-H]^-$.

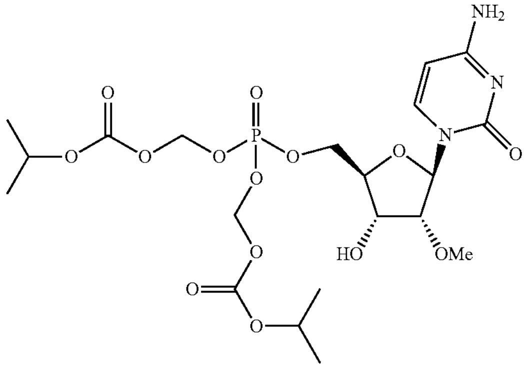
Step B: 3'-Deoxycytidine-5'-[bis-(S-pivaloyl-2-thioethyl)phosphate]

This compound was synthesized following the similar synthetic procedure used for the synthesis of Example 80 starting with N^4-(4,4'-dimethoxytrityl)-3'-deoxycytidine (0.3 g, 0.57 mmol). Other reagents used for the synthesis were 1H-tetrazole (0.04 g, 0.57 mmol), acetonitrile (4 mL), bis-(S-pivaloyl-2-thioethyl)-N,N-diisopropylphosphoramidite (0.52 g, 1.14 mmol) and 3-chloroperbenzoic acid (0.2 g, 1.14 mmol, 57-80%) in CH_2Cl_2 (3.6 mL). The product was isolated in 22% yield (0.073 g) after HPLC purification.

^{1}H NMR (200 MHz, DMSO-d_6): δ 1.17 (s, 18H), 1.84 (m, 2H), 3.11 (t, 4H, J=6.4 Hz), 3.93-4.31 (m, 8H), 4.39 (m, 1H), 5.55 (d, 1H, J=4.2 Hz), 5.67 (dd, 2H, J=7.4 and 1.8 Hz), 7.1 (bs, 2H), 7.56 (d, 1H, J=7.4 Hz); ^{13}P NMR (CD_3CN): δ −0.71; MS (AP-ES) m/z 596.1 $[M+H]^+$; HRMS (MALDI) Calcd for $C_{23}H_{38}N_3O_9PS_2Na$ 618.1679 found 618.1600.

EXAMPLE 83

2'-O-Methylcytidine-5'-[bis(isopropyloxycarbonyloxymethyl)]phosphate



Phosphonomethoxy nucleoside analogs are discussed in C. R. Wagner, V. V. Iyer, and E. J. McIntee, "Pronucleotides: Toward the In Vivo Delivery of Antiviral and Anticancer Nucleotides," *Med. Res. Rev.*, 20:1-35 (2000), which is incorporated by reference herein in its entirety. They are also disclosed U.S. Pat. Nos. 5,922,695; 5,977,089; 6,043,230; and 6,069,249, the contents of each of which are incorporated by reference herein in their entirety.

Step A: iso-Propyl chloromethyl carbonate

This was prepared according to *Antiviral Chemistry & Chemotherapy* 8: 557 (1997).

Step B: 2'-O-Methylcytidine-5'-phosphate

This intermediate was prepared as described in *Tetrahedron Lett.* 50: 5065 (1967).

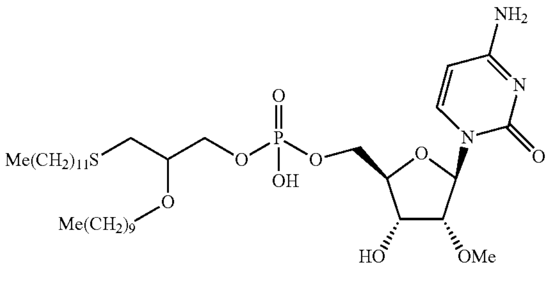
Step C: 2'-O-Methylcytidine-5'-[bis(isopropyloxycarbonyloxy methyl)]phosphate

2'-O-Methylcytidine-5'-phosphate (0.4 g, 1.19 mmol) was dried over P_2O_5 in vacuum overnight at 40° C. It was then suspended in anhydrous DMF (4 mL). To this mixture was added diisopropylethylamine (0.86 mL, 4.92 mmol) and isopropyl chloromethyl carbonate (1.56 g, 7.34 mmol). The mixture was heated at 50° C. for 1 h. The reaction mixture was then allowed to come to room temperature. The reaction mixture was stirred at room temperature for 48 h and then filtered. The filtrate was diluted with water (100 mL) and extracted with CH_2Cl_2 (3×50 mL). The organic phase was dried over anhydrous Na_2SO_4 and evaporated. The residue was dissolved in 20% MeOH in water and purified by HPLC on C-18 column (Luna C18,250×2.12 mm, A=water, B=acetonitrile 20 to 10% B in 65 mL, flow 10 mL min^{-1}, λ 260 nm) to give the title compound (2.5 mg).

^{13}P NMR (CD_3CN): δ −3.09; MS (AP-ES) m/z 570.1 $[M+H]^+$.

EXAMPLE 84

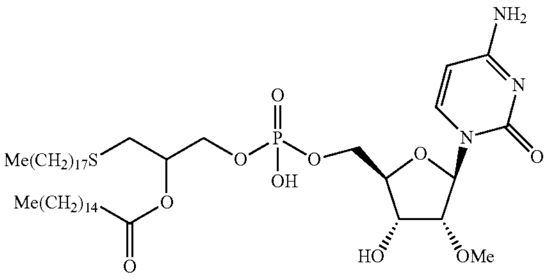
2'-O-Methylcytidine-5'-[(2-decyloxy-3-dodecylthio-1-propyl)phosphate]



The procedure is described for similar nucleoside analogs in German Patent 408366 (1992) and *J. Acquired Immune Defic. Syndr.* 2000, 23, 227. The reaction of the appropriately protected 2'-O-methylcytidine with (2-decyloxy-3-dodecylthio-1-propyl)phosphate [prepared by the reaction of 2-decyloxy-3-dodecylthio-1-propanol with $POCl_3$ in ether in presence of triethylamine] under refluxing conditions in a toluene-ether mixture furnishes the desired compound.

EXAMPLE 85

2'-O-Methylcytidine-5'-[rac-(3-octadecylthio-2-palmitoyloxy-1-propyl)phosphate]



This compound is synthesized by the reaction of 2'-O-methylcytidine-5'-monophosphoromorpholidate with rac-1-S-octadecyl-2-O-palmitoyl-1-thioglycerol in pyridine following the similar procedure described for AZT and ddC in *J. Med. Chem.* 39: 1771 (1996).

EXAMPLE 86

Nucleoside 5'-Triphosphates

The nucleoside 5'-triphosphates of the present invention were prepared according to the general procedures described in *Chem. Rev.* 100: 2047 (2000).

EXAMPLE 87

Purification and Purity Analysis of Nucleoside 5'-Triphosphates

Triphosphates were purified by anion exchange (AX) chromatography using a 30×100 mm Mono Q column (Pharmacia) with a buffer system of 50 mM Tris, pH 8. Elution gradients were typically from 40 mM NaCl to 0.8 M NaCl in two column volumes at 6.5 mL/min. Appropriate fractions from anion exchange chromatography were collected and desalted by reverse-phase (RP) chromatography using a Luna C18 250×21 mm column (Phenomenex) with a flow rate of 10 ml/min. Elution gradients were generally from 1% to 95% methanol in 14 min at a constant concentration of 5 mM triethylammonium acetate (TEAA).

Mass spectra of the purified triphosphates were determined using on-line HPLC mass spectrometry on a Hewlett-Packard (Palo Alto, Calif.) MSD 1100. A Phenomenex Luna (C18(2)), 150×2 mm, plus 30×2 mm guard column, 3-μm particle size was used for RP HPLC. A 0 to 50% linear gradient (15 min) of acetonitrile in 20 mM TEAA (triethylammonium acetate) pH 7 was performed in series with mass spectral detection in the negative ionization mode. Nitrogen gas and a pneumatic nebulizer were used to generate the electrospray. The mass range of 150-900 was sampled. Molecular masses were determined using the HP Chemstation analysis package.

The purity of the purified triphosphates was determined by analytical RP and AX HPLC. RP HPLC with a Phenomonex Luna or Jupiter column (250×4.6 mm), 5-μm particle size was typically run with a 2-70% acetonitrile gradient in 15 min in

100 mM TEAA, pH 7. AX HPLC was performed on a 1.6×5 mm Mono Q column (Pharmacia). Triphosphates were eluted with a gradient of 0 to 0.4 M NaCl at constant concentration of 50 mM Tris, pH 8. Purity of the triphosphates was generally >80%.

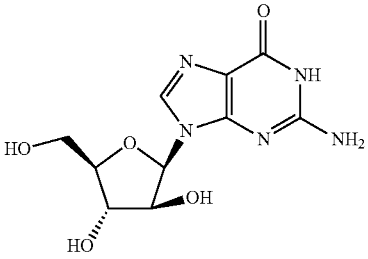
EXAMPLE 88

Nucleoside 5'-Monophosphates

The nucleoside 5'-monophosphates of the present invention were prepared according to the general procedure described in *Tetrahedron Lett.* 50: 5065 (1967).

EXAMPLE 89

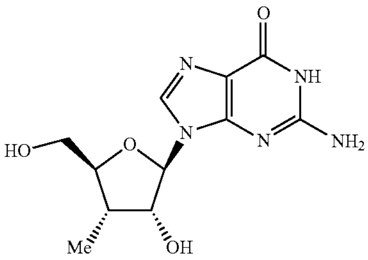
2-Amino-9-(β-D-arabinofuranosyl)-9H-purin-6(1H)-one



This compound was obtained from commercial sources.

EXAMPLE 90

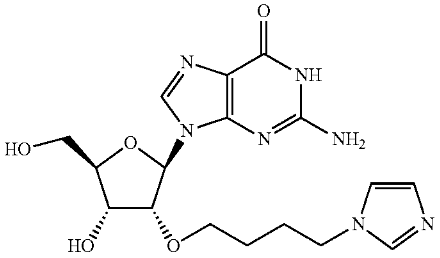
3'-Deoxy-3'-methylguanosine



This compound was prepared following procedures described in U.S. Pat. No. 3,654,262 (1972).

EXAMPLE 91

2'-O-[4-(Imidazolyl-1)butyl]guanosine



Step A: 2'-O-[4-(Imidazolyl-1)butyl]-2-aminoadenosine

A solution 2-aminoadenosine (7.36 g, 26 mmol) in dry DMF (260 mL) was treated portionwise with 60% NaH (3.92 g, 1000 mmol). After 1 hr., a solution of bromobutylimidazole (9.4 g, 286 mmol) in DMF (20 ml) was added. After 16 hrs., the solution was conc. in vacuo, partitioned between H_2O/EtOAc and separated. The aqueous layer was evaporated, and the residue was chromatographed on silica gel ($CHCl_3$/MeOH) to afford the title nucleoside as a white solid; yield 4.2 g.

^1H NMR (DMSO-d_6): δ 1.39 (t, 2H), 1.67 (t, 2H), 3.3-3.7 (m, 4H), 3.93 (m, 3H), 4.29 (m, 2H), 4.40 (d, 1H), 5.50 (5, 1H), 5.72 (d, 1H), 5.82 (bs, 2H), 6.72 (bs, 2H), 6.86 (s, 1H), 7.08 (s, 1H), 7.57 (s, 1H). 7.91 (s, 1H).

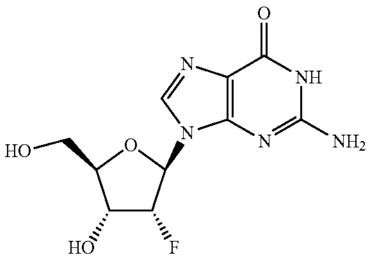
Step B: 2'-O-[4-(Imidazolyl-1)butyl]guanosine

A mixture of the intermediate from Step A (3.2 g, 8 mmol) in H_2O (200 mL), DMSO (10 mL), trisodium phosphate (10 g), and adenosine deaminase (0.3 g) was stirred at room temperature and pH 7. The solution was filtered and then evaporated. The resulting solid was crystallized from EtOAc/MeOH to afford the title compound as a white solid; yield 2.6

^1H NMR (DMSO-d_o): δ 1.39 (t, 2H), 1.67 (t, 2H), 3.3-3.7 (m, 4H), 3.93 (m, 3H), 4.29 (m, 2H), 5.10 (t, 1H), 5.20 (d, 1H), 5.79 (d, 1H), 6.50 (bs, 2H), 6.86 (s, 1H), 7.08 (s, 1H), 7.57 (s, 1H) 7.9 (s, 1H).

EXAMPLE 92

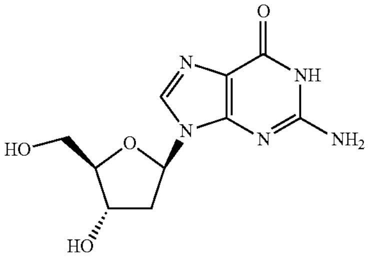
2'-Deoxy-2'-fluoroguanosine



This compound was prepared following the conditions described in *Chem. Pharm. Bull.* 29: 1034 (1981). g.

EXAMPLE 93

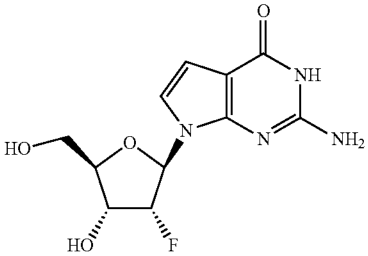
2'-Deoxyguanosine



This compound was obtained from commercial sources.

EXAMPLE 94

2-Amino-7-(2-deoxy-2-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



Step A: 2-Amino-4-chloro-7-(2,3,5-tri-O-benzyl-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]-pyrimidine

To a suspension of 2-amino-4-chloro-1H-pyrrolo[2,3-d] pyrimidine [*Liebigs Ann. Chem.* 1: 137 (1983)] (3.03 g, 18 mmol) in anhydrous MeCN (240 mL), powdered KOH (85%; 4.2 g, 60 mmol) and tris[2-(2-methoxyethoxy)-ethyl]amine (0.66 mL, 2.1 mmol) were added and the mixture was stirred at room temperature for 10 min. Then a solution of 2,3,5-tri-O-benzyl-D-arabinofuranosyl bromide [prepared from corresponding 1-O-p-nitrobenzoate (11.43 g, 20.1 mmol) according to Seela et al., *J. Org. Chem.* (1982), 47, 226] in MeCN (10 mL) was added and stirring continued for another 40 min. Solid was filtered off, washed with MeCN (2×25 mL) and combined filtrate evaporated. The residue was purified on a silica gel column with a solvent system of hexanes/EtOAc:7/1, 6/1 and 5/1. Two main zones were separated. From the more rapidly migrating zone was isolated the α anomer (0.74 g) and from the slower migrating zone the desired β anomer (4.01 g).

Step B: 2-Amino-7-(β-D-arabino furanosyl)-4-chloro-7H-pyrrolo[2,3-d]pyrimidine

To a solution of the compound from Step A (4.0 g, 7 mmol) in CH_2Cl_2 (150 ml) at −78° C. was added a solution of 1.0 M BCl_3 in CH_2Cl_2 (70 mL, 70 mmol) during 45 min. The mixture was stirred at −78° C. for 3 h and at −20° C. for 2.5 h. MeOH—CH_2Cl_2 (70 mL, 1:1) was added to the mixture, which was then stirred at −20° C. for 0.5 h and neutralized with conc. aqueous NH_3 at 0° C. The mixture was stirred at room temperature for 10 min. and then filtered. The solid was washed with MeOH—CH_2Cl_2 (70 mL, 1:1) and the combined filtrate evaporated. The residue was purified on a silica gel column with a solvent system of CH_2Cl_2/MeOH: 20/1 to give the desired nucleoside (1.18 g) as a white solid.

Step C: 2-Amino-7-[3,5-O-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)-β-D-arabinofuranosyl]-4-chloro-7H-pyrrolo[2,3-d]pyrimidine

The compound from Step B (0.87 g, 2.9 mmol) and imidazole (0.43 g, 5.8 mmol were dissolved in DMF (3.5 mL). 1,3-Dichloro-1,1,3,3-tetraisopropyldisiloxane (1.0 mL) was added to the solution. The reaction mixture was stirred at room temperature for 1 h and then evaporated. The residue was partitioned between CH_2Cl_2 (150 mL) and water (30 mL). The layers were separated. The organic layer was dried (Na_2SO_4) and evaporated. The residue was purified on a silica gel column with a solvent system of hexanes/EtOAc:7/1 and 5/1 to give the title compound (1.04 g).

Step D: 2-Amino-7-[2-O-acetyl-3,5-O-(1,1,3,3-tetraisopropyldisiloxane-1,3-diyl)-β-D-arabinofuranosyl]-4-chloro-7H-pyrrolo[2,3-d]pyrimidine

A mixture of the compound from Step C (0.98 g, 1.8 mmol) in MeCN (12 mL), Et_3N (0.31 mL) Ac_2O (0.21 mL) and DMAP (5 mg, 0.25 eq.) was stirred at room temperature for 5 h and then evaporated. The oily residue was dissolved in EtOAc (200 mL), washed with water (2×20 mL), dried (Na_2SO_4) and evaporated to yield pure title compound (1.12 g).

Step E: 2-Amino-7-[2-O-acetyl-β-D-arabinofuranosyl]-4-chloro-7H-pyrrolo[2,3-d]pyrimidine

To an ice-cold solution of the compound from Step D (0.95 g, 1.63 mmol) in THF (10 mL) and AcOH (0.19 mL) was added dropwise 1.0 M tetrabutylammonium fluoride solution in THF (3.4 mL) and stirred at 0° C. for 15 min. The solution was concentrated and the oily residue applied onto a silica gel column packed in CH_2Cl_2 and eluted with CH_2Cl_2/MeOH: 50/1, 25/1 and 20/1. Appropriate fractions were pooled and evaporated to give the title nucleoside (0.56 g) as a white solid.

Step F: 2-Amino-7-[2-O-acetyl-3,5-di-O-(tetrahydro-2-pyranyl)-β-D-arabinofuranosyl]-4-chloro-7H-pyrrolo[2,3-d]pyrimidine

To a solution of the compound from Step E (0.5 g, 1.46 mmol) in CH_2Cl_2 (10 mL) and 3,4-dihydro-2-H-pyrane (0.67 mL) was added dropwise TMSI (30 μL, 0.2 mmol). The reaction mixture was stirred at room temperature for 1 h and then evaporated. The oily residue was purified on a silica gel column packed in a solvent system of hexanes/EtOAc/Et_3N: 75/25/1 and eluted with a solvent system of hexanes/EtOAc: 3/1. The fractions containing the product were collected and evaporated to give the desired compound (0.60 g).

Step G: 2-Amino-7-[3,5-di-O-(tetrahydro-2-pyranyl)-β-D-arabinofuranosyl]-4-chloro-7H-pyrrolo[2,3-d]pyrimidine

A mixture of the compound from Step F (0.27 g, 0.53 mmol) and methanolic ammonia (saturated at 0° C.; 10 mL) was kept overnight at 0° C. Evaporation of the solvent yielded the desired compound (0.25 g).

Step H: 2-Amino-7-[2-deoxy-2-fluoro-3,5-di-O-(tetrahydro-2-pyranyl)-β-D-ribofuranosyl]-4-chloro-7H-pyrrolo[2,3-d]pyrimidine

To a solution of the compound from Step G (0.24 g, 0.51 mmol) in CH_2Cl_2 (5 mL) and pyridine (0.8 mL) at −60° C. was added diethylaminosulfur trifluoride (DAST; 0.27 mL) dropwise under Ar. The solution was stirred at −60° C. for 0.5 h, at 0° C. overnight and at room temperature for 3 h. The mixture was diluted with CH_2Cl_2 (25 mL) and poured into saturated aqueous $NaHCO_3$ (15 mL). The organic layer was washed with water (10 mL), dried (Na_2SO_4) and evaporated. The residue was purified on a silica gel column with a solvent system of hexanes/EtOAc:5/1 to give the title compound (45 mg) as a pale yellow foam.

Step I: 2-Amino-7-(2-deoxy-2-fluoro-β-D-ribofuranosyl)-4-chloro-7H-pyrrolo[2,3-d]-pyrimidine

A solution of the compound from Step H (40 mg. 0.08 mmol) in EtOH (2 mL) was stirred with pyridinium p-toluenesulfonate (40 mg, 0.16 mmol) at 60° C. for 3 h. The mixture was then evaporated and the residue purified on a silica gel column with a solvent system of hexanes/EtOAc: 1/1 and 1/2 to give the desired compound (24 mg).

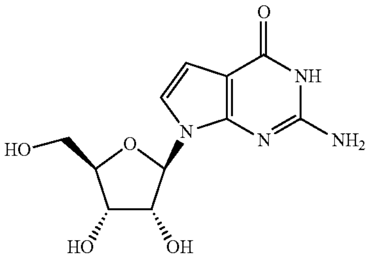
Step J: 2-Amino-7-(2-deoxy-2-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one

A mixture of the compound from Step I (4 mg, 0.08 mmol) in 2N aqueous NaOH (1.2 mL) was stirred at reflux temperature for 1.5 h. The solution was cooled in an ice-bath, neutralized with 2 N aqueous HCl and evaporated to dryness. The residue was suspended in MeOH, mixed with silica gel and evaporated. The solid residue was placed onto a silica gel column (packed in a solvent system of CH_2Cl_2/MeOH: 10/1) which was eluted with a solvent system of CH_2Cl_2/MeOH: 10/1. The fractions containing the product were collected and evaporated to dryness to yield the title compound (20 mg) as a white solid. ^1H NMR (CD_3OD): δ 3.73, 3.88 (2dd, 2H, J=12.4, 3.8, 2.6 Hz), 4.01 (m, 1H), 4.47 (ddd, 1H J=16.5, 6.6 Hz), 5.14 (ddd, 1H, J=5.3, 4.7 Hz), 6.19 (dd, 1H, J=17.8, 3.0 Hz), 6.39 (d, 1H, J=3.6 Hz), 6.95 (d, 1H).

^{19}F NMR (CD_3OD): δ −206.53 (dt).

EXAMPLE 95

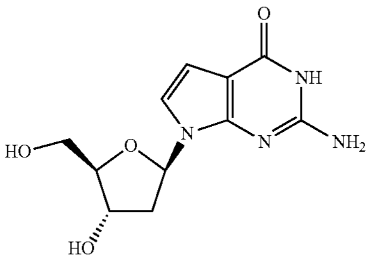
2-Amino-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



This compound was prepared following the procedures described in *J. Chem. Soc. Perkin Trans.* 1, 2375 (1989).

EXAMPLE 96

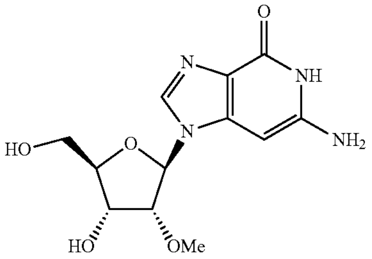
2-Amino-7-(2-deoxy-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



This compound was prepared following the procedures in *Tetrahedron Lett.* 28: 5107 (1987).

EXAMPLE 97

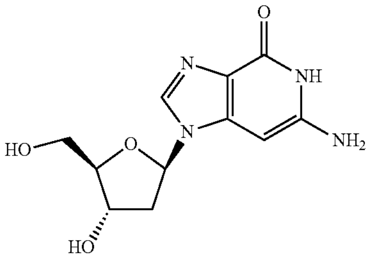
6-Amino-1-(2-O-methyl-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one



This compound was prepared in a manner similar to the preparation of 2-amino-7-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one (Example 23).

EXAMPLE 98

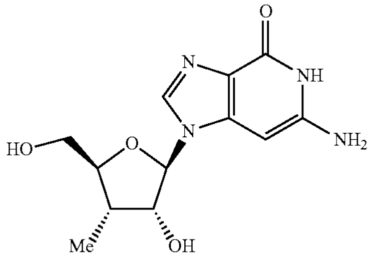
6-Amino-1-(2-deoxy-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one



This compound was prepared following the procedures described in *J. Med. Chem.* 26: 286 (1983).

EXAMPLE 99

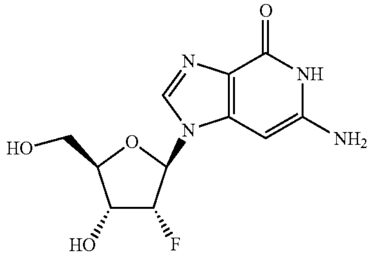
6-Amino-1-(3-deoxy-3-methyl-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one



This compound was prepared in a manner similar to the preparation of 2-amino-7-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-c]pyrimidin-4(3H)-one (Example 23).

EXAMPLE 100

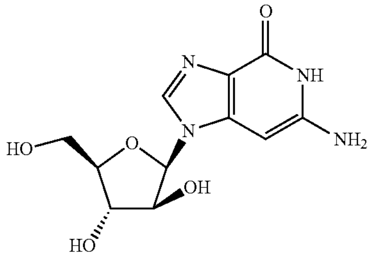
6-Amino-1-(2-deoxy-2-fluoro-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one



This compound was prepared in a manner similar to the preparation of 2-amino-7-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-7H-pyrrolo[2,3-c]pyrimidin-4(3H)-one (Example 23).

EXAMPLE 101

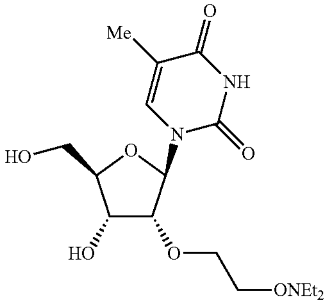
6-Amino-1-(β-D-arabinofuranosyl)-1H-imidazo[4,5-c]pyridin-4(5H)-one



A preparation of this compound is given in Eur. Pat. Appln. 43722 A1 (1982).

EXAMPLE 102

2'-O-[2-(N,N-diethylaminooxy)ethyl]-5-methyluridine



Step A: 5'-O-tert-Butyldiphenylsilyl-2'-O-(2-hydroxyethyl)-5-methyluridine

In a 2 L stainless steel, unstirred pressure reactor was added borane in tetrahydrofuran (1.0 M, 2.0 eq, 622 mL). In the fume hood and with manual stirring, ethylene glycol (350 mL, excess) was added cautiously at first until the evolution of hydrogen gas subsided. 5'-O-tert-Butyldiphenylsilyl-O^2-2'-anhydro-5-methyluridine (149 g, 0.311 mol) and sodium bicarbonate (0.074 g) were added with manual stirring. The reactor was sealed and heated in an oil bath until an internal temperature of 160° C. was reached and then maintained for 16 h (pressure<100 psig). The reaction vessel was cooled to ambient and opened. The reaction mixture was concentrated under reduced pressure (10 to 1 mm Hg) in a warm water bath (40-100° C.) with the more extreme conditions used to remove the ethylene glycol. The residue was purified by column chromatography (2 kg silica gel, ethyl acetate:hexanes gradient 1:1 to 4:1). The appropriate fractions were combined, stripped and dried to product as white crisp foam (84 g), contaminated starting material (17.4 g) and pure reusable starting material (20 g). TLC and NMR were consistent with 99% pure product.

^{1}H NMR (DMSO-d_6): δ 1.05 (s, 9H), 1.45 (s, 3H), 3.5-4.1 (m, 8H), 4.25 (m, 1H), 4.80 (t, 1H), 5.18 (d, 2H), 5.95 (d, 1H), 7.35-7.75 (m, 11H), 11.42 (s, 1H).

Step B: 2'-O-[2-(2-phthalimidoxy)ethyl]-5'-t-butyldiphenylsilyl-5-methyluridine

5'-O-tert-Butyldiphenylsilyl-2'-O-(2-hydroxyethyl)-5-methyluridine (20 g, 36.98 mmol) was mixed with triphenylphosphine (11.63 g, 44.36 mmol) and N-hydroxyphthalimide (7.24 g, 44.36 mmol). It was then dried over P_2O_5 under high vacuum for two days at 40° C. The reaction mixture was flushed with argon and dry THF (369.8 mL) was added to get a clear solution. Diethyl azodicarboxylate (6.98 mL, 44.36 mmol) was added dropwise to the reaction mixture. The rate of addition was maintained such that resulting deep red coloration is just discharged before adding the next drop. After the addition was complete, the reaction was stirred for 4 h. By that time TLC showed the completion of the reaction (ethyl acetate/hexane, 60:40). The solvent was evaporated under vacuum. Residue obtained was placed on a flash silica gel column and eluted with ethyl acetate-hexane (60:40) to give the title compound as a white foam (21.8 g).

^{1}H NMR (DMSO-d_6): δ 11.32 (s, 1H), 7.82 (m, 4H), 7.6-7.65 (m, 5H), 7.34-7.46 (m, 6H), 5.90 (d, 1H, J=6 Hz), 5.18 (d, J=5.6 Hz), 4.31 (bs, 2H), 4.25 (m, 1H), 4.09 (t, 1H, J=5.6 Hz), 3.81-3.94 (m, 5H), 1.44 (s, 3H), 1.1 (s, 9H); ^{13}C NMR ($CDCl_3$): δ 11.8, 19.40, 26.99, 62.62, 68.36, 68.56, 77.64, 83.04, 84.14, 87.50, 110.93, 123.59, 127.86, 129.89, 132.45, 134.59, 134.89, 135.17, 150.50, 163.63, 163.97; MS [FAB] m/z 684 $[M-H]^-$.

Step C: 5'-O-tert-Butyldiphenylsilyl-2'-O-[2-(acetaldoximinooxy)ethyl]-5-methyluridine

2'-O-[2-(2-Phthalimidoxy)ethyl]-5'-t-butyldiphenylsilyl-5-methyluridine (10 g, 14.6 mmol) was dissolved in CH_2Cl_2 (146 mL) and cooled to −10° C. in an isopropanol-dry ice bath. To this methylhydrazine (1.03 mL, 14.6 mmol) was added dropwise. Reaction mixture was stirred at −10° C. to 0° C. for 1 h. A white precipitate formed and was filtered and washed thoroughly with CH_2Cl_2 (ice cold). The filtrate was evaporated to dryness. Residue was dissolved in methanol (210 mL) and acetaldehyde (0.89 mL, 16 mmol) was added and stirred at room temperature for 12 h. Solvent was removed in vacuo and residue was purified by silica gel column chromatography using and ethyl acetate/hexane (6:4) as solvent system to yield the title compound (4.64 g).

^{1}H NMR (DMSO-d_6): δ 1.02 (s, 9H), 1.44 (s, 3H), 1.69 (dd, 3H, J=5.6 Hz), 3.66 (m, 1H), 3.76 (m, 2H), 3.94 (m, 2H), 4.05 (s, 2H), 4.15 (m, 1H), 4.22 (m, 1H), 5.18 (d, 1H, J=6.0 Hz), 5.9 (dd, 1H, J=4.4 Hz), 7.36 (m, 1H), 7.40 (m, 7H), 7.63 (m, 5H), 11.38 (s, 1H), ^{13}C NMR ($CDCl_3$): δ 11.84, 15.05, 19.38, 26.97, 63.02, 68.62, 70.26, 71.98, 72.14, 82.72, 84.34, 87.02, 111.07, 127.89, 130.02, 134.98, 135.13, 135.42, 147.85, 150.51, 164.12; HRMS (FAB) Calcd for $C_{30}H_{39}N_3O_7SiNa^{\oplus}$ 604.2455, found 604.2471.

Step D: 5'-O-tert-Butyldiphenylsilyl-2'-O-[2-(N,N-diethylaminooxy)ethyl]-5-methyluridine

5'-O-tert-Butyldiphenylsilyl-2'-O-[2-(acetaldoximinooxy)ethyl]-5-methyluridine (4.5 g, 7.74 mmol) was dissolved in 1M pyridinium p-toluenesulfonate (PPTS) in MeOH (77.4 mL). It was then cooled to 10° C. in an ice bath. To this mixture $NaBH_3CN$ (0.97 g, 15.5 mmol) was added and the mixture was stirred at 10° C. for 10 minutes. Reaction mixture was allowed to come to room temperature and stirred for 4 h. Solvent was removed in vacuo to give an oil. Diluted the oil with ethyl acetate (100 mL), washed with water (75 mL), 5% $NaHCO_3$ (75 mL) and brine (75 mL). The organic phase was dried over anhydrous Na_2SO_4 and evaporated. Residue obtained was dissolved in 1M PPTS in MeOH (77.4 mL), acetaldehyde (0.48 mL, 8.52 mmol) was added and stirred at ambient temperature for 10 minutes. Then reaction mixture was cooled to 10° C. in an ice bath and $NaBH_3CN$ (0.97 g, 15.50 mmol) was added and stirred at 10° C. for 10 minutes. Reaction mixture was allowed to come to room temperature and stirred for 4 h. Solvent was removed in vacuo to get an oil. The oil was dissolved in ethyl acetate (100 mL), washed with water (75 mL), 5% $NaHCO_3$ (75 mL) and brine (75 mL). The organic phase was dried over anhydrous Na_2SO_4 and evaporated to dryness. The residue obtained was purified by silica gel column chromatography and eluted with CH_2Cl_2/MeOH/NEt_3, 94:5:1 to give title compound (3.55 g) as a white foam.

^{1}H NMR (DMSO-d_6): δ 0.95 (t, 6H, J=7.2 Hz), 1.03 (s, 9H), 1.43 (s, 3H), 2.58 (q, 4H, J=7.2 Hz), 3.59 (m, 1H), 3.73 (m, 3H), 3.81 (m, 1H), 3.88 (m, 1H), 3.96 (m, 2H), 4.23 (m, 1H), 5.21 (d, 1H, J=5.6 Hz), 5.95 (d, 1H, J=6.4 Hz), 7.43 (m, 7H), 7.76 (m, 4H), 11.39 (s, 1H); ^{13}C NMR ($CDCl_3$): δ 11.84, 19.35, 26.97, 52.27, 63.27, 68.81, 70.27, 72.27, 82.64, 84.47, 86.77, 111.04, 127.87, 130.01, 135.11, 135.41, 141.32, 150.48, 164.04; HRMS (FAB), Calcd for $C_{32}H_{45}N_3O_7SiCs^{\oplus}$, 744.2081, found 744.2067.

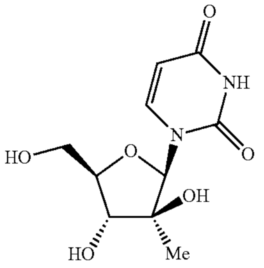
Step E: 2'-O-[2-(N,N-diethylaminooxy)ethyl]-5-methyluridine

A mixture of triethlyamine trihydrogenfluoride (4.39 mL, 26.81 mmol) and triethylamine (1.87 mL, 13.41 mmol) in THF (53.6 mL) was added to 5'-O-tert-butyldiphenylsilyl-2'-O-[2-(N,N-diethylaminooxy)ethyl]-5-methyluridine (3.28 g, 5.36 mmol). The reaction mixture was stirred at room temperature for 18 h. Solvent was removed in vacuo. The residue was placed on a silica gel column and eluted with CH_2Cl_2/MeOH/NEt_3, 89:10:1, to yield the title compound (1.49 g).

^{1}H NMR (DMSO-d_6): δ 0.97 (t, 6H, J=7.2 Hz), 1.75 (s, 3H), 2.58 (q, 4H, J=7.2 Hz), 3.55 (m, 4H), 3.66 (m, 2H), 3.83 (bs, 1H), 3.95 (t, 1H, J=5.6 Hz), 4.11 (q, 1H, J=4.8 Hz and 5.6 Hz), 5.05 (d, 1H, J=5.6 Hz), 5.87 (d, 1H, J=6.0 Hz), 7.75 (s, 1H), 11.31 (s, 1H); ^{13}C NMR ($CDCl_3$): δ 11.75, 12.27, 52.24, 61.31, 68.86, 70.19, 72.25, 81.49, 85.10, 90.29, 110.60, 137.79, 150.57, 164.37; HRMS (FAB) Calcd for $C_{16}H_{28}N_3O_7^{\oplus}$ 374.1927, found 374.1919.

EXAMPLE 103

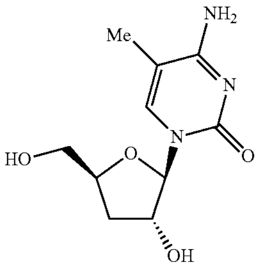
1-(2-C-Methyl-β-D-arabinofuranosyl)uracil



This compound was prepared following the procedures described in *Chem. Pharm. Bull.* 35: 2605 (1987).

EXAMPLE 104

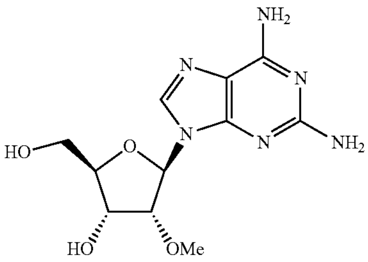
5-Methyl-3'-deoxycytidine



This compound was prepared following the procedures described in *Chem. Pharm. Bull.* 30: 2223 (1982).

EXAMPLE 105

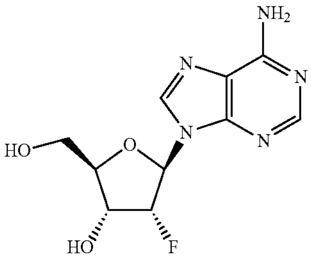
2-Amino-2'-O-methyladenosine



This compound was obtained from commercial sources.

EXAMPLE 106

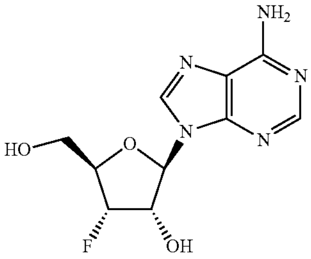
2'-Deoxy-2'-fluoroadenosine



This compound was obtained from commercial sources.

EXAMPLE 107

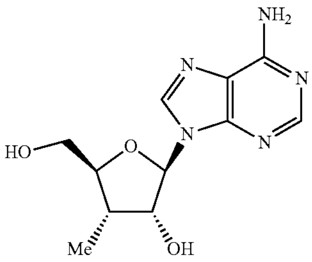
3'-Deoxy-3'-fluoroadenosine



This compound was prepared following the procedures described in *Nucleosides Nucleotides* 10: 719 (1991).

EXAMPLE 108

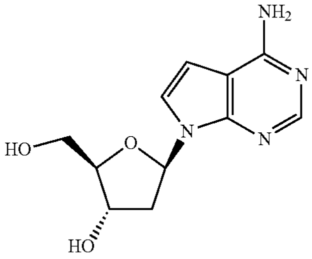
3'-Deoxy-3'-methyladenosine



This compound was prepared following the procedures described in *J. Med. Chem.* 19: 1265 (1976).

EXAMPLE 109

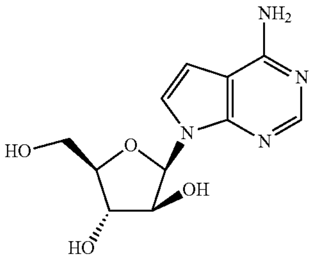
2-Amino-7-(2-deoxy-β-D-ribofuranosyl)-7H-pyrrolo [2,3-d]pyrimidine



This compound was prepared following the procedures described in *J. Am. Chem. Soc.* 106: 6379 (1984).

EXAMPLE 110

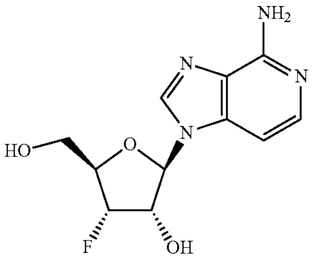
4-Amino-7-(β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



This compound is described in U.S. Pat. No. 4,439,604, which is incorporated by reference herein in its entirety.

EXAMPLE 111

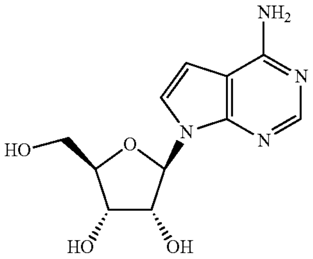
4-Amino-1-(3-deoxy-3-fluoro-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridine



This compound can be prepared readily by the similar method described for the preparation of Example 24 except the nucleobase is 3-deazaadenine.

EXAMPLE 112

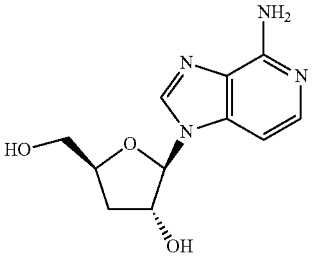
4-Amino-7-(β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine (tubercidin)



This compound was obtained from commercial sources.

EXAMPLE 113

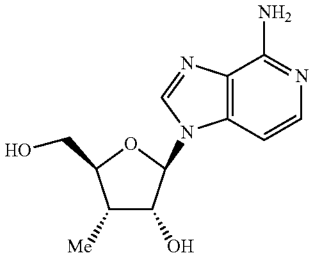
4-Amino-1-(3-deoxy-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridine



This compound is described in *Acta Crystallogr., Sect. C: Cryst. Struct. Commun*. C43: 1790 (1987).

EXAMPLE 114

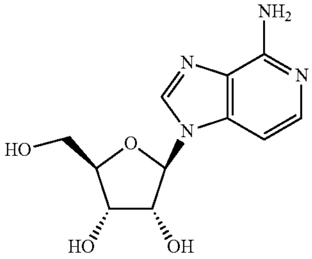
4-Amino-1-(3-deoxy-3-methyl-β-D-ribofuranosyl)-1H-imidazo[4,5-c]pyridine



The procedure described earlier for Example 23 is used to synthesize this example by reacting the appropriately substituted 3-C-methyl-sugar intermediate with a protected 3-deazaadenine derivative.

EXAMPLE 115

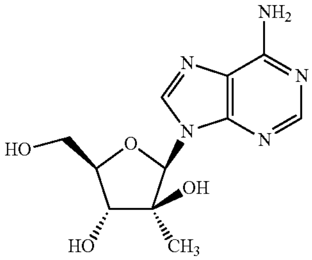
4-Amino-1-β-D-ribofuranosyl-1H-imidazo[4,5-c]pyridine



This compound was obtained from commercial sources.

EXAMPLE 116

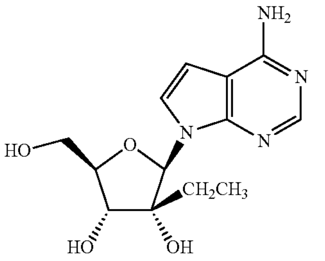
9-(2-C-Methyl-β-D-arabinofuranosyl)adenine



This compound is prepared from 4-amino-9-(3,5-bis-O-tert-butyldimethylsilyl-β-D-erythro-pentofuran-2-ulosyl) purine (*J. Med. Chem.* 1992, 35, 2283) by reaction with MeMgBr and deprotection as described in Example 61.

EXAMPLE 117

4-Amino-7-(2-C-ethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



Step A: 3,5-Bis-O-(2,4-dichlorophenylmethyl)-2-C-ethyl-1-O-methyl-α-D-ribofuranose

To diethyl ether (300 mL) at −78° C. was slowly added EtMgBr (3.0 M, 16.6 mL) and then dropwise the compound from Step B of Example 62 (4.80 g, 10.0 mmol) in anhydrous Et_2O (100 mL). The reaction mixture was stirred at −78° C. for 15 min, allowed to warm to −15° C. and stirred for another 2 h, and then poured into a stirred mixture of water (300 mL) and Et_2O (600 mL). The organic phase was separated, dried ($MgSO_4$), and evaporated in vacuo. The crude product was purified on silica gel using ethyl acetate/hexane (1:2) as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (3.87 g) as a colorless oil.

Step B: 4-Chloro-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C-ethyl-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine

To a solution of the compound from Step A (1.02 mg, 2.0 mmol) in dichloromethane (40 mL) was added HBr (5.7 M in acetic acid) (1.75 mL, 10.0 mmol) dropwise at 0° C. The resulting solution was stirred at rt for 2 h, evaporated in vacuo and co-evaporated twice from toluene (10 mL). The oily residue was dissolved in acetonitrile (10 mL) and added to a vigorously stirred mixture of 4-chloro-1H-pyrrolo[2,3-d]pyrimidine (307 mg, 2.0 mmol), potassium hydroxide (337 mg, 6.0 mmol) and tris[2-(2-methoxyethoxy)ethyl]amine (130 mg, 0.4 mmol) in acetonitrile (10 mL). The resulting mixture was stirred at room temperature overnight, and then poured into a stirred mixture of saturated ammonium chloride (100 mL) and ethyl acetate (100 mL). The organic layer was separated, washed with brine (100 mL), dried over $MgSO_4$, filtered and evaporated in vacuo. The crude product was purified on silica gel using ethyl acetate/hexane (1:2) as eluent to give the desired product (307 mg) as a colorless foam.

Step C: 4-Chloro-7-(2-C-ethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

To a solution of the compound from Step B (307 mg, 0.45 mmol) in dichloromethane (8 mL) was added boron trichloride (1M in dichloromethane) (4.50 mL, 4.50 mmol) at −78° C. The mixture was stirred at −78° C. for 1 h, then at −10° C. for 3 h. The reaction was quenched by addition of methanol/dichloromethane (1:1) (10 mL), stirred at −15° C. for 30 min, and neutralized by addition of aqueous ammonium hydroxide. The mixture was evaporated in vacuo and the resulting oil purified on silica gel using methanol/dichloromethane (1:9) as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (112 mg) as a colorless foam.

Step D: 4-Amino-7-(2-C-ethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

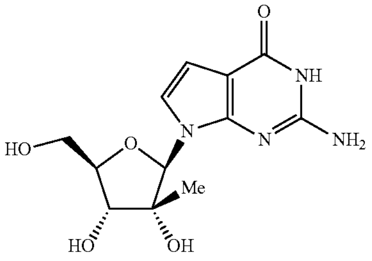
To the compound from Step C (50 mg, 0.16 mmol) was added saturated ammonia in methanol (4 mL). The mixture was stirred at 75° C. for 72 h in a closed container, cooled and evaporated in vacuo. The crude mixture was purified on silica gel using methanol/dichloromethane (1:9) as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (29 mg) as a colorless powder.

^1H NMR (200 MHz, DMSO-d_6): δ 0.52 (t, 3H), 1.02 (m, 2H), 4.01-3.24 (m, 6H), 5.06 (m, 1H), 6.01 (s, 1H), 6.51 (d, 1H), 6.95 (s br, 2H), 6.70 (d, 1H), 7.99 (s, 1H).

LC-MS: Found: 295.2 (M+H^+); calc. for $C_{13}H_{18}N_4O_4+H^+$: 295.14.

EXAMPLE 118

2-Amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



Step A: 2-Amino-4-chloro-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

To an ice-cold solution of product from Step C of Example 62 (1.27 g, 2.57 mmol) in CH_2Cl_2 (30 mL) was added HBr (5.7 M in acetic acid; 3 mL) dropwise. The reaction mixture was stirred at room temperature for 2 h, concentrated in vacuo and co-evaporated with toluene (2×15 mL). The resulting oil was dissolved in MeCN (15 mL) and added dropwise into a well-stirred mixture of 2-amino-4-chloro-7H-pyrrolo[2,3-d]pyrimidine [for preparation see *Heterocycles* 35: 825 (1993)] (433 mg, 2.57 mmol), KOH (85%, powdered) (0.51 g, 7.7 mmol), tris-[2-(2-methoxyethoxy)ethyl]amine (165 μL, 0.51 mmol) in acetonitrile (30 mL). The resulting mixture was stirred at rt for 1 h, filtered and evaporated. The residue was purified on a silica gel column using hexanes/EtOAc, 5/1, 3/1 and 2/1 as eluent to give the title compound as a colorless foam (0.65 g).

Step B: 2-Amino-4-chloro-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

To a solution of the product from Step A (630 mg, 1.0 mmol) in CH_2Cl_2 (20 mL) at −78° C. was added boron trichloride (1M in CH_2Cl_2) (10 mL, 10 mmol). The mixture was stirred at −78° C. for 2 h, then at −20° C. for 2.5 h. The reaction was quenched with CH_2Cl_2/MeOH (1:1) (10 mL), stirred at −20° C. for 0.5 h, and neutralized at 0° C. with aqueous ammonia. The solid was filtered, washed with CH_2Cl_2/MeOH (1:1) and the combined filtrate evaporated in vacuo. The residue was purified on a silica gel column with CH_2Cl_2/MeOH, 50/1 and 20/1 as eluent to give the title compound as a colorless foam (250 mg).

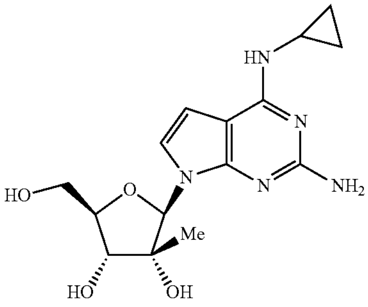
Step C: 2-Amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one

A mixture of product from Step B (90 mg, 0.3 mmol) in aqueous NaOH (2N, 9 mL) was heated at reflux temperature for 5 h, then neutralized at 0° C. with 2 N aqueous HCl and evaporated to dryness. Purification on a silica gel column with CH_2Cl_2/MeOH, 5/1 as eluent afforded the title compound as a white solid (70 mg).

^{1}H NMR (200 MHz, CD_3OD): δ 0.86 (s, 3H), 3.79 (m 1H), 3.90-4.05 (m, 3H), 6.06 (s, 1H), 6.42 (d, J=3.7 Hz, 1H), 7.05 (d, J=3.7 Hz, 1H).

EXAMPLE 119

2-Amino-4-cyclopropylamino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidie

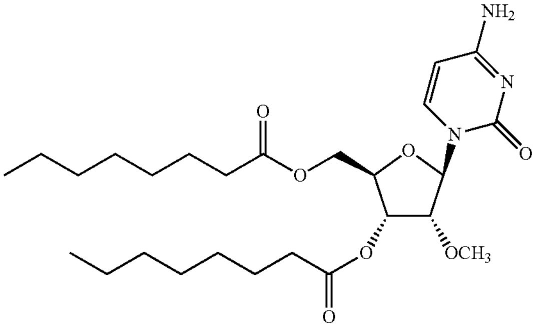


A solution of 2-amino-4-chloro-7-(2-C-methyl-θ-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine (Example 118, Step B) (21 mg, 0.07 mmol) in cyclopropylamine (0.5 mL) was heated at 70° C. for two days, then evaporated to an oily residue and purified on a silica gel column with CH_2Cl_2/MeOH, 20/1, as eluent to give the title compound as a white solid (17 mg).

^{1}H NMR (200 MHz, CD_3CN): δ 0.61 (m, 2H), 0.81 (m, 2H), 0.85 (s, 3H), 2.83 (m, 1H), 3.74-3.86 (m, 1H), 3.93-4.03 (m, 2H), 4.11 (d, J=8.9 Hz, 1H), 6.02 (s, 1H), 6.49 (d, J=3.7 Hz, 1H), 7.00 (d, J=3.7 Hz, 1H).

EXAMPLE 120

3',5'-Bis-[O-(1-oxooctyl)]-2'-O-methylcytidine



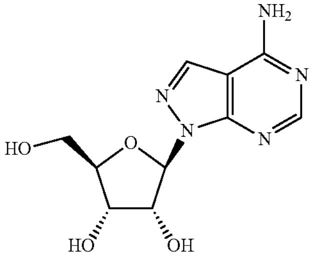
1,3-Dicyclohexylcarbodiimide (21.48 g, 104 mmol) was dissolved in anhydrous dichloromethane (100 mL). To the solution was added octanoic acid (5.49 mL, 34.5 mmol, made anhydrous by keeping over molecular sieves, 4 A° overnight at room temperature), and the resulting reaction mixture was stirred under argon atmosphere for 6 h. The white precipitate which formed was filtered, and the filtrate was concentrated under reduced pressure. The residue obtained was dissolved in anhydrous pyridine and added to N^4-(4,4'-dimethoxytrityl)-2'-O-methylcytidine (0.43 g, 0.77). DMAP (0.09 g, 0.77 mmol) was added and the resulting mixture was stirred at room temperature under argon atmosphere for 12 h. The solvent was removed under reduced pressure and the residue obtained was dissolved in ethyl acetate (100 mL). The organic phase was washed with aqueous sodium bicarbonate (5%, 50 mL), dried over anhydrous Na_2SO_4 and concentrated under reduced pressure. The residue was purified by flash silica gel column chromatography and eluted with 5% MeOH in dichloromethane. The product obtained was dissolved in a mixture of acetic acid: MeOH:H_2O (20 mL, 3:6:1). The resulting mixture was heated at 50° C. for 24 h. The solvent was removed under reduced pressure. The residue obtained was purified by flash silica gel column chromatography and eluted with dichloromethane containing 0 to 5% of MeOH to give the title compound (0.22 g).

^{1}H NMR (200 MHz, DMSO-d_6) δ 0.83 (m, 6H), 1.23 (br s, 16H), 1.51 (m, 4H), 2.33 (m, 4H), 3.26 (s, 3H), 4.06 (t, J=5.2 Hz, 1H), 4.21 (m, 3H), 5.11 (t, J=5.2 Hz, 1H), 5.75 (d, J=7.4 Hz, 1H), 5.84 (d, J=4.8 Hz, 1H), 7.26 (br s, 2H), 7.61 (d, J=7.4 Hz, 1H).

MS (ES): m/z 510.3 $[M+H]^+$; HRMS (FAB) Calcd for $C_{26}H_{44}N_3O_7$: 510.3179; found 510.3170.

EXAMPLE 121

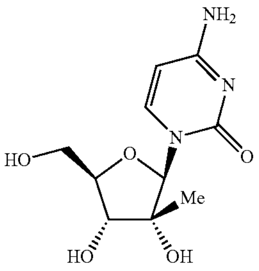
4-Amino-1-(θ-D-ribofuranosyl)-1H-pyrazolo[3,4-d]pyrimidine



This compound was prepared following procedures described in *Nucleic Acids Res.,* 11: 871-872 (1983).

EXAMPLE 122

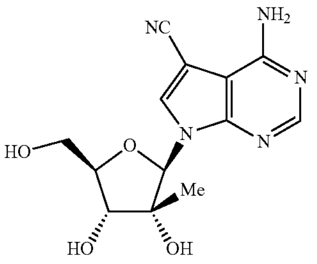
2'-C-Methyl-cytidine



This compound was prepared following procedures described in L. Beigelman et al., *Carbohyd. Res.* 166: 219-232 (1987) or X-Q Tang, et al., *J. Org. Chem.* 64: 747-754 (1999).

EXAMPLE 123

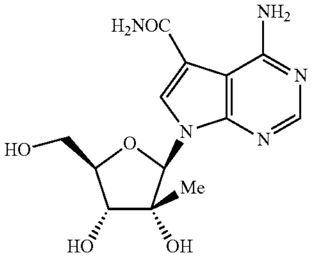
4-Amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carbonitrile



This compound was prepared following procedures described by Y. Murai et al. in *Heterocycles* 33: 391-404 (1992).

EXAMPLE 124

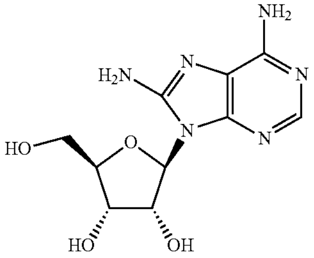
4-Amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carboxamide



This compound was prepared following procedures described by Y. Murai et al. in *Heterocycles* 33: 391-404 (1992).

EXAMPLE 125

8-Aminoadenosine



This compound was prepared following the procedure described in M. Ikehara and S. Yamada, *Chem. Pharm. Bull.,* 19: 104 (1971).

EXAMPLE 126

Mass Spectral Characterization of Nucleoside 5'-Triphosphates

Mass spectra of nucleoside 5'-triphosphates were determined as described in Example 87. Listed in the following table are the calculated and experimental masses for the nucleoside 5'-triphosphates prepared according to the procedures of Example 86. The example numbers correspond to the parent nucleoside of the nucleoside 5'-triphosphate.

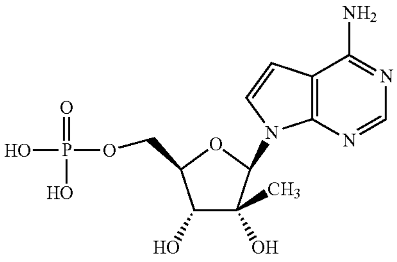
Example	Calculated	Found
1	507.0	506.9
2	525.0	524.9
5	537.0	537.0
6	539.0	539.0
7	565.0	565.0
8	547.0	546.9
9	550.0	550.0
10	506.0	505.9
11	536.0	535.9
12	536.0	536.0
13	561.0	560.9
14	550.0	550.0
15	524.0	524.0
16	522.0	521.9
17	547.0	546.9
18	536.0	536.0
20	531.0	530.9
21	522.0	522.0
22	536.0	536.0
23	506.0	506.1
24	524.0	524.0
25	508.0	508.0
26	508.0	508.0
27	552.0	552.0
28	506.0	506.0
29	579.0	578.9
30	582.0	582.0
31	568.0	567.9
32	554.0	553.9
33	540.0	539.9
34	554.0	553.9
35	568.0	567.9

-continued

Example	Calculated	Found
36	541.0	541.0
37	565.0	564.9
38	542.0	541.9
39	554.0	553.9
41	481.0	481.0
42	467.0	467.0
43	485.0	484.8
46	482.0	482.0
47	486.0	485.8
48	482.0	482.0
49	554.0	554.0
51	468.0	468.1
52	521.0	521.0
53	491.0	491.2
55	584.9	585.1
56	521.0	521.2
58	506.0	506.0
61	520.0	519.9
62	520.0	520.0
63	547.0	547.0
64	533.0	533.0
65	549.0	549.0
67	551.0	551.0
68	515.0	514.9
69	520.0	520.1
71	490.0	489.9
89	523.0	522.9
90	521.0	520.9
91	645.1	645.0
94	524.0	523.9
95	522.0	521.8
98	536.0	535.9
99	520.0	520.0
102	613.1	613.0
103	498.0	497.9
104	481.0	481.0
105	536.0	536.2
106	509.0	508.9
108	505.0	505.0
112	506.0	506.1
113	490.0	490.0
117	534.0	534.0
118	536.0	536.0

EXAMPLE 127

[4-Amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]-pyrimidine]-5'-monophosphate

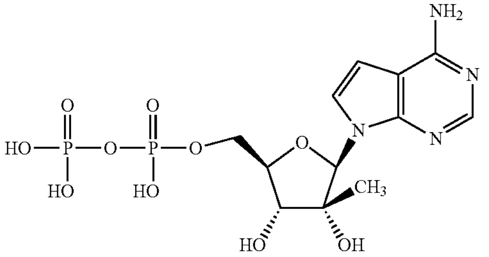


To the compound from Step F of Example 62 (14 mg, 0.05 mmol) (dried by coevaporation with pyridine and several times with toluene) was added trimethyl phosphate (0.5 mL). The mixture was stirred overnight in a sealed container. It was then cooled to 0° C. and phosphorous oxychloride (0.0070 mL, 0.075 mmol) was added via a syringe. The mixture was stirred for 3 h at 0° C., then the reaction was quenched by addition of tetraethylammonium bicarbonate (TEAB) (1M) (0.5 mL) and water (5 mL). The reaction mixture was purified and analyzed according to the procedure described in Example 87.

Electron spray mass spectrum (ES-MS): Found: 359.2 (M–H$^+$), calc. for $C_{12}H_{17}N_4O_7P$—H$^+$: 359.1.

EXAMPLE 128

[4-Amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]-pyrimidine]-5'-diphosphate

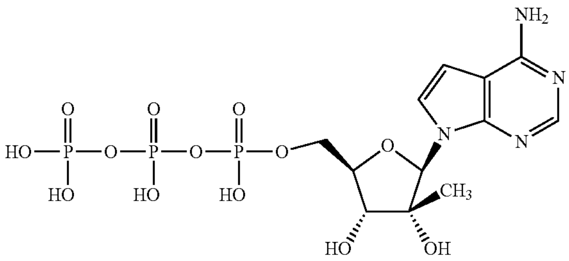


To the compound from Step F of Example 62 (56 mg, 0.20 mmol) (dried by coevaporation with pyridine and several times with toluene) was added trimethyl phosphate (stored over sieves) (1.0 mL). The mixture was stirred overnight in a sealed container. It was then cooled to 0° C. and phosphorous oxychloride (0.023 mL, 0.25 mmol) was added via a syringe. The mixture was stirred for 2 h at 0° C., then tributylamine (0.238 mL, 1.00 mmol) and tributylammonium phosphate (generated from phosphoric acid and tributylamine in pyridine, followed by repeated azeotropic evaporation with pyridine and acetonitrile) (1.0 mmol in 3.30 mL acetonitrile) was added. The mixture was stirred for an additional 30 min at 0° C., the sealed vial was then opened and the reaction quenched by addition of TEAB (1M) (1.0 mL) and water (5 mL). The reaction mixture was purified and analyzed according to the procedure described in Example 87.

ES-MS: Found: 439.0 (M–H$^+$), calc. for $C_{12}H_{18}N_4O_{10}P_2$—H$^+$: 439.04.

EXAMPLE 129

[4-Amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]-pyrimidine]-5'-triphosphate



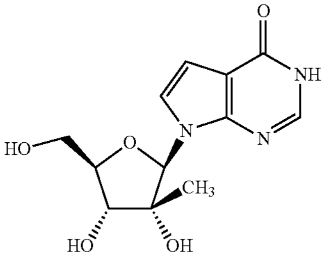
To the compound from Step F of Example 62 (20 mg, 0.07 mmol) (dried by coevaporation with pyridine and several times with toluene) was added trimethyl phosphate (stored over sieves) (0.4 mL). The mixture was stirred overnight in a sealed container. It was then cooled to 0° C. and phosphorous oxychloride (0.0070 mL, 0.075 mmol) was added via syringe.

The mixture was stirred for 3 h at 0° C., then tributylamine (0.083 mL, 0.35 mmol), tributylammonium pyrophosphate (0.35 mmol, 127 mg) and acetonitrile (stored over sieves) (0.25 mL) were added. The mixture was stirred for an additional 30 min at 0° C., the sealed vial was then opened and the reaction quenched by addition of TEAB (1M) (0.5 mL) and water (5 mL). The reaction mixture was purified and analyzed according to the procedure described in Example 87.

ES-MS: Found: 519.0 (M−H$^+$), calc. for $C_{12}H_{19}N_4O_{13}P_3$—H$^+$: 519.01.

EXAMPLE 130

7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one

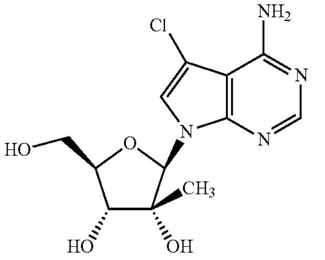


To the compound from Step E of Example 62 (59 mg, 0.18 mmol) was added aqueous sodium hydroxide (1M). The mixture was heated to reflux for 1 hr, cooled, neutralized with aqueous HCl (2M) and evaporated in vacuo. The residue was purified on silica gel using dichloromethane/methanol (4:1) as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (53 mg) as a colorless oil.

^{1}H NMR (CD_3CN): δ 0.70 (s, 3H), 3.34-4.15 (overlapping m, 7H), 6.16 (s, 1H), 6.57 (d, 3.6 Hz, 1H), 7.37 (d, 3.6 Hz, 1H), 8.83 (s, 1H).

EXAMPLE 131

4-Amino-5-chloro-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



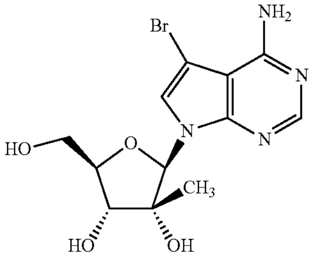
To a pre-cooled solution (0° C.) of the compound from Step F of Example 62 (140 mg, 0.50 mmol) in DMF (2.5 mL) was added N-chlorosuccinimide (0.075 g, 0.55 mmol) in DMF (0.5 mL) dropwise. The solution was stirred at rt for 1 h and the reaction quenched by addition of methanol (4 mL) and evaporated in vacuo. The crude product was purified on silica gel using methanol/dichloromethane (1:9) as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (55 mg) as a colorless solid.

^{1}H NMR (CD_3CN): δ 0.80 (s, 3H), 3.65-4.14 (overlapping m, 7H), 5.97 (s br, 2H), 6.17 (s, 1H), 7.51 (s, 1H), 8.16 (s, 1H).

ES-MS: Found: 315.0 (M+H$^+$), calc. for $C_{12}H_{15}ClN_4O_4$+H$^+$: 315.09.

EXAMPLE 132

4-Amino-5-bromo-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



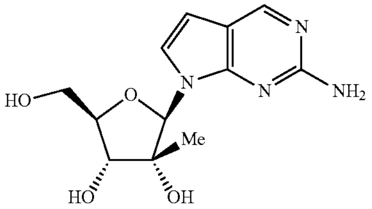
To a pre-cooled solution (0° C.) of the compound from Step F of Example 62 (28 mg, 0.10 mmol) in DMF (0.5 mL) was added N-bromosuccinimide (0.018 g, 0.10 mmol) in DMF (0.5 mL) dropwise. The solution was stirred at 0° C. for 20 min, then at rt for 10 min. The reaction was quenched by addition of methanol (4 mL) and evaporated in vacuo. The crude product was purified on silica gel using methanol/dichloromethane (1:9) as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (13.0 mg) as a colorless solid.

^{1}H NMR (CD_3CN): δ 0.69 (s, 3H), 3.46-4.00 (overlapping m, 7H), 5.83 (s br, 2H), 6.06 (s, 1H), 7.45 (s, 1H), 8.05 (s, 1H).

ES-MS: Found: 359.1 (M+H$^+$), calc. for $C_{12}H_{15}BrN_4O_4$+H$^+$: 359.04.

EXAMPLE 133

2-Amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

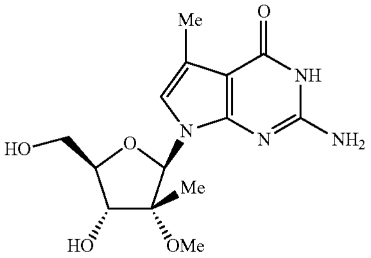


A mixture of 2-amino-4-chloro-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-c]pyrimidine (Example 118, Step B) (20 mg, 0.07 mmol) in EtOH (1.0 mL), pyridine (0.1 mL) and 10% Pd/C (6 mg) under H_2 (atmospheric pressure) was stirred overnight at room temperature. The mixture was filtered through a Celite pad which was thoroughly washed with EtOH. The combined filtrate was evaporated and purified on a silica gel column with CH_2Cl_2/MeOH, 20/1 and 10/1, as eluent to give the title compound as a white solid (16 mg).

^{1}H NMR (200 MHz, CD_3OD): δ 0.86 (s, 3H, 2'C-Me), 3.82 (dd, $J_{5',4'}$=3.6 Hz, $J_{5',5''}$=12.7 Hz, 1H, H-5'), 3.94-4.03 (m, 2H, H-5', H-4'), 4.10 (d, $J_{3'4'=8.8}$ Hz, 1H, H-3'), 6.02 (s, 1H, H-1'), 6.41 (d, $J_{5,6}$=3.8 Hz, 1H, H-5), 7.39 (d, 1H, H-6), 8.43 (s, 1H, H-4). ES MS: 281.4 (MH^+).

EXAMPLE 134

2-Amino-5-methyl-7-(2-C,2-O-dimethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one



Step A: 2-Amino-4-chloro-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C-methyl-β-D-ribofuranosyl]-5-methyl-7H-pyrrolo[2,3-d]pyrimidine

To an ice-cold solution of the product from Step C of Example 62 (1.57 g, 3.16 mmol) in CH_2Cl_2 (50 mL) was added HBr (5.7 M in acetic acid; 3.3 mL) dropwise. The reaction mixture was stirred at 0° C. for 1 h and then at room temperature for 2 h, concentrated in vacuo and co-evaporated with toluene (2×20 mL). The resulting oil was dissolved in MeCN (20 mL) and added dropwise to a solution of the sodium salt of 2-amino-4-chloro-5-methyl-1H-pyrrolo[2,3-d]pyrimidine in acetonitrile [generated in situ from 2-amino-4-chloro-5-methyl-1H-pyrrolo[2,3-d]pyrimidine [for preparation, see *Liebigs Ann. Chem.* 1984: 708-721] (1.13 g, 6.2 mmol) in anhydrous acetonitrile (150 mL), and NaH (60% in mineral oil, 248 mg, 6.2 mmol), after 2 h of vigorous stirring at rt]. The combined mixture was stirred at rt for 24 h and then evaporated to dryness. The residue was suspended in water (100 mL) and extracted with EtOAc (300+150 mL). The combined extracts were washed with brine (100 mL), dried over Na_2SO_4, filtered and evaporated. The crude product was purified on a silica gel column (5×7 cm) using ethyl acetate/hexane (0 to 30% EtOAc in 5% step gradient) as the eluent. Fractions containing the product were combined and evaporated in vacuo to give the desired product (0.96 g) as a colorless foam.

Step B: 2-Amino-4-chloro-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C,2-O-dimethyl-β-D-ribofuranosyl]-5-methyl-7H-pyrrolo[2,3-d]pyrimidine

To an ice-cold mixture of the product from Step A (475 mg, 0.7 mmol) in THF (7 mL) was added NaH (60% in mineral oil, 29 mg) and stirred at 0° C. for 0.5 h. Then MeI (48 μL) was added and reaction mixture stirred at rt for 24 h. The reaction was quenched with MeOH and the mixture evaporated. The crude product was purified on a silica gel column (5×3.5 cm) using hexane/ethyl acetate (9/1, 7/1, 5/1 and 3/1) as eluent. Fractions containing the product were combined and evaporated to give the desired compound (200 mg) as a colorless foam.

Step C: 2-Amino-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C,2-O-dimethyl-β-D-ribofuranosyl]-5-methyl-7H-pyrrolo[2,3-d]pyrimidine-4(3H)-one

A mixture of the product from Step B (200 mg, 0.3 mmol) in 1,4-dioxane (15 mL) and aqueous NaOH (2N, 15 mL) in a pressure bottle was heated overnight at 135° C. The mixture was then cooled to 0° C., neutralized with 2N aqueous HCl and evaporated to dryness. The crude product was suspended in MeOH, filtered, and the solid thoroughly washed with MeOH. The combined filtrate was concentrated, and the residue purified on a silica gel column (5×5 cm) using CH_2Cl_2/MeOH (40/1, 30/1 and 20/1) as eluent to give the desired compound (150 mg) as a colorless foam.

Step D: 2-Amino-5-methyl-7-(2-C,2-O-dimethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one

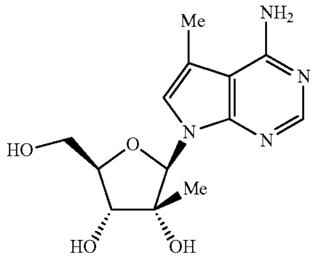
A mixture of the product from Step C (64 mg, 0.1 mmol) in MeOH (5 mL) and Et_3N (0.2 mL) and 10% Pd/C (24 mg) was hydrogenated on a Parr hydrogenator at 50 psi at r.t. for 1.5 days, then filtered through a Celite pad which was thoroughly washed with MeOH. The combined filtrate was evaporated and the residue purified on a silica gel column (3×4 cm) with CH_2Cl_2/MeOH (30/1, 20/1) as eluent to yield 2-amino-5-methyl-7-(5-O-benzyl-2-C,2-O-dimethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidin-4(3H)-one. The compound (37 mg) was further hydrogenated in EtOH (2 mL) with 10% Pd/C and under atmospheric pressure of hydrogen. After stirring 2 days at r.t., the reaction mixture was filtered through Celite, the filtrate evaporated and the crude product purified on a silica gel column (1×7 cm) with CH_2Cl_2/MeOH (30/1, 20/1 and 10/1) as eluent to yield the title compound (12 mg) after freeze-drying.

^{1}H NMR (200 MHz, CD_3OD): δ 0.81 (s, 3H, 2'C-Me), 2.16 (d, $J_{H\text{-}6,C5\text{-}Me}$=1.3 Hz, 3H, C5-Me), 3.41 (s, 3H, 2'-OMe), 3.67 (dd, $J_{5'4'}$=3.4 Hz, $J_{5',5''}$=12.6 Hz, 1H, H-5'), 3.81-3.91 (m, 3H, H-5", H-4', H-3'), 6.10 (s, 1H, H-1'), 6.66 (d, 1H, H-6).

ES MS: 323.3 $(M-H)^+$.

EXAMPLE 135

4-Amino-5-methyl-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



Step A: 4-Chloro-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C-methyl-β-D-ribofuranosyl]-5-methyl-7H-pyrrolo[2,3-d]pyrimidine

To an ice-cold solution of the product from Step C of Example 62 (1.06 g, 2.1 mmol) in CH_2Cl_2 (30 mL) was added HBr (5.7 M in acetic acid; 2.2 mL) dropwise. The reaction mixture was stirred at 0° C. for 1 h and then at room temperature for 2 h, concentrated in vacuo and co-evaporated with toluene (2×15 mL). The resulting oil was dissolved in MeCN (10 mL) and added dropwise into a solution of the sodium salt of 4-chloro-5-methyl-1H-pyrrolo[2,3-d]pyrimidine in acetonitrile [generated in situ from 4-chloro-5-methyl-1H-pyrrolo[2,3-d]pyrimidine [for preparation, see *J. Med. Chem.* 33: 1984 (1990)] (0.62 g, 3.7 mmol) in anhydrous acetonitrile (70 mL), and NaH (60% in mineral oil, 148 mg, 3.7 mmol), after 2 h of vigorous stirring at rt]. The combined mixture was stirred at rt for 24 h and then evaporated to dryness. The residue was suspended in water (100 mL) and extracted with EtOAc (250+100 mL). The combined extracts were washed with brine (50 mL), dried over Na_2SO_4, filtered and evaporated. The crude product was purified on a silica gel column (5×5 cm) using hexane/ethyl acetate (9/1, 5/1, 3/1) gradient as the eluent. Fractions containing the product were combined and evaporated in vacuo to give the desired product (0.87 g) as a colorless foam.

Step B: 4-Chloro-5-methyl-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

To a solution of the compound from Step A (0.87 g, 0.9 mmol) in dichloromethane (30 mL) at −78° C. was added boron trichloride (1M in dichloromethane, 9.0 mL, 9.0 mmol) dropwise. The mixture was stirred at −78° C. for 2.5 h, then at −30° C. to −20° C. for 3 h. The reaction was quenched by addition of methanol/dichloromethane (1:1) (9 mL) and the resulting mixture stirred at −15° C. for 30 min., then neutralized with aqueous ammonia at 0° C. and stirred at rt for 15 min. The solid was filtered and washed with CH_2Cl_2/MeOH (1/1, 50 mL). The combined filtrate was evaporated, and the residue was purified on a silica gel column (5×5 cm) using CH_2Cl_2 and CH_2Cl_2/MeOH (40/1 and 30/1) gradient as the eluent to furnish the desired compound (0.22 g) as a colorless foam.

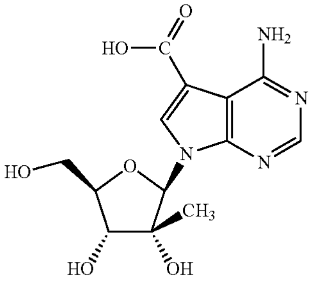
Step C: 4-Amino-5-methyl-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

To the compound from Step B (0.2 g, 0.64 mmol) was added methanolic ammonia (saturated at 0° C.; 40 mL). The mixture was heated in a stainless steel autoclave at 100° C. for 14 h, then cooled and evaporated in vacuo. The crude mixture was purified on a silica gel column (5×5 cm) with CH_2Cl_2/MeOH (50/1, 30/1, 20/1) gradient as eluent to give the title compound as a white solid (0.12 g).

^{1}H NMR (DMSO-d_6): δ 0.60 (s, 3H, 2'C-Me), 2.26 (s, 3H, 5C-Me), 3.52-3.61 (m, 1H, H-5'), 3.70-3.88 (m, 3H, H-5", H-4', H-3'), 5.00 (s, 1H, 2'-OH), 4.91-4.99 (m, 3H, 2'-OH, 3'-OH, 5'-OH), 6.04 (s, 1H, H-1'), 6.48 (br s, 2H, NH_2), 7.12 (s, 1H, H-6), 7.94 (s, 1H, H-2). ES MS: 295.2 (MH^+).

EXAMPLE 136

4-Amino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine-5-carboxylic acid



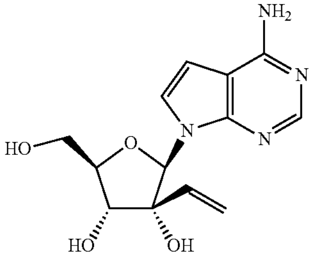
The compound of Example 123 (0.035 g, 0.11 mmol) was dissolved in a mixture of aqueous ammonia (4 mL, 30 wt %) and saturated methanolic ammonia (2 mL), and a solution of H_2O_2 in water (2 mL, 35 wt %) was added. The reaction mixture was stirred at room temperature for 18 h. Solvent was removed under reduced pressure, and the residue obtained was purified by HPLC on a reverse phase column (Altech Altima C-18, 10×299 mm, A=water, B=acetonitrile, 10 to 60% B in 50 min, flow 2 mL/min) to yield the title compound (0.015 g, 41%) as a white solid.

^{1}H NMR (CD_3OD): δ 0.85 (s, 3H, Me), 3.61 (m, 1H), 3.82 (m, 1H) 3.99-4.86 (m, 2H), 6.26 (s, 1H), 8.10 (s, 2H) 8.22 (s, 1H); ^{13}C NMR (CD_3OD): 20.13, 61.37, 73.79, 80.42, 84.01, 93.00, 102.66, 112.07, 130.07, 151.40, 152.74, 159.12, 169.30.

HRMS (FAB) Calcd for $C_{13}H_{17}N_4O_6^+$325.1148, found 325.1143.

EXAMPLE 137

4-Amino-7-(2-C-vinyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



Step A: 3,5-Bis-O-(2,4-dichlorophenylmethyl)-2-C-vinyl-1-O-methyl-α-D-ribofuranose

Cerium chloride heptahydrate (50 g, 134.2 mmol) was finely crushed in a pre-heated mortar and transferred to a round-bottom flask equipped with a mechanical stirrer. The flask was heated under high vacuum overnight at 160° C. The vacuum was released under argon and the flask was cooled to room temperature. Anhydrous THF (300 mL) was cannulated into the flask. The resulting suspension was stirred at room temperature for 4 h and then cooled to −78° C. Vinylmagnesium bromide (1M in THF, 120 mL, 120 mmol) was added and stirring continued at −78° C. for 2 h. To this suspension was added a solution of 3,5-bis-O-(2,4-dichlorophenylmethyl)-1-O-methyl-α-D-erythro-pentofuranose-2-ulose (14 g, 30 mmol) [from Example 2, Step B] in anhydrous THF (100 mL), dropwise with constant stirring. The reaction was stirred at −78° C. for 4 h. The reaction was quenched with saturated ammonium chloride solution and allowed to come to room temperature. The mixture was filtered through a celite pad and the residue washed with Et_2O (2×500 mL). The organic layer was separated and the aqueous layer extracted with Et_2O (2×200 mL). The combined organic layers were dried over anhydrous Na_2SO_4 and concentrated to a viscous yellow oil. The oil was purified by flash chromatography (SiO_2, 10% EtOAc in hexanes). The title compound (6.7 g, 13.2 mmol) was obtained as a pale yellow oil.

Step B: 4-Chloro-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C-vinyl-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine

To a solution of the compound from Step A (6.4 g, 12.6 mmol) in anhydrous dichloromethane (150 mL) at −20° C. was added HBr (30% solution in AcOH, 20 mL, 75.6 mmol) dropwise. The resulting solution was stirred between −10° C. and 0° C. for 4 h, evaporated in vacuo and co-evaporated with anhydrous toluene (3×40 mL). The oily residue was dissolved in anhydrous acetonitrile (100 mL) and added to a solution of the sodium salt of 4-chloro-1H-pyrrolo[2,3-d]pyrimidine (5.8 g, 37.8 mmol) in acetonitrile (generated in situ as described in Example 62) at −20° C. The resulting mixture was allowed to come to room temperature and stirred at room temperature for 24 h. The mixture was then evaporated to dryness, taken up in water and extracted with EtOAc (2×300 mL). The combined extracts were dried over Na_2SO_4, filtered and evaporated. The crude mixture was purified by flash chromatography (SiO_2, 10% EtOAc in hexanes) and the title compound (1.75 g) isolated as a white foam.

Step C: 4-Amino-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C-vinyl-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine

The compound from Step B (80, mg) was dissolved in the minimum amount of 1,4-dioxane and placed in a stainless steel bomb. The bomb was cooled to −78° C. and liquid ammonia was added. The bomb was sealed and heated at 90° C. for 24 h. The ammonia was allowed to evaporate and the residue concentrated to a white solid which was used in the next step without further purification.

Step D: 4-Amino-7-(2-C-vinyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

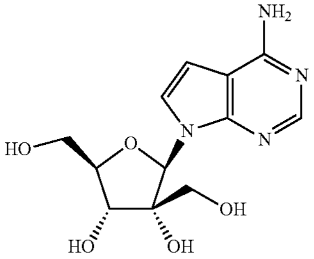
To a solution of the compound from Step C (60 mg) in dichloromethane at −78° C. was added boron trichloride (1M in dichloromethane) dropwise. The mixture was stirred at −78° C. for 2.5 h, then at −30° C. to −20° C. for 3 h. The reaction was quenched by addition of methanol/dichloromethane (1:1) and the resulting mixture stirred at −15° C. for 0.5 h, then neutralized with aqueous ammonia at 0° C. and stirred at room temperature for 15 min. The solid was filtered and washed with methanol/dichloromethane (1:1). The combined filtrate was evaporated and the residue purified by flash chromatography (SiO_2, 10% methanol in EtOAc containing 0.1% triethylamine). The fractions containing the product were evaporated to give the title compound as a white solid (10 mg).

^1H NMR (DMSO-d_6): δ 3.6 (m, 1H, H-5'), 3.8 (m, 1H, H-5"), 3.9 (m d, 1-H, H-4'), 4.3 (t, 1H, H-3'), 4.8-5.3 (m, 6H, $CH{=}CH_2$, 2'-OH, 3'-OH, 5'-OH) 6.12 (s, 1H, H-1'), 6.59 (d, 1H, H-5), 7.1 (br s, 1H, NH_2), 7.43 (d, 1H, H-6), 8.01 (s, 1H, H-2).

ES-MS: Found: 291.1 (M−H^-); calc. for $C_{13}H_{16}N_4O_4$—H^-: 291.2.

EXAMPLE 138

4-Amino-7-(2-C-hydroxymethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



Step A: 4-Chloro-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C-hydroxymethyl-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine

To a solution of the compound from Example 137, Step B (300 mg, 0.48 mmol) in 1,4-dioxane (5 mL) were added N-methylmorpholine-N-oxide (300 mg, 2.56 mmol) and osmium tetroxide (4% solution in water, 0.3 mL). The mixture was stirred in the dark for 14 h. The precipitate was removed by filtration through a celite plug, diluted with water (3×), and extracted with EtOAc. The EtOAc layer was dried over Na_2SO_4 and concentrated in vacuo. The oily residue was taken up in dichloromethane (5 mL) and stirred over $NaIO_4$ on silica gel (3 g, 10% $NaIO_4$) for 12 h. The silica gel was removed by filtration and the residue was evaporated and taken up in absolute ethanol (5 mL). The solution was cooled in an ice bath and sodium borohydride (300 mg, 8 mmol) was added in small portions. The resulting mixture was stirred at room temperature for 4 h and then diluted with EtOAc. The organic layer was washed with water (2×20 mL), brine (20 mL) and dried over Na_2SO_4. The solvent was evaporated and the residue purified by flash chromatography (SiO_2, 2:1 hexanes/EtOAc) to give the title compound (160 mg, 0.25 mmol) as white flakes.

Step B: 4-Amino-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C-hydroxymethyl-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine

The compound from Step A (150 mg, 0.23 mmol) was dissolved in the minimum amount of 1,4-dioxane (10 mL) and placed in a stainless steel bomb. The bomb was cooled to −78° C. and liquid ammonia was added. The bomb was sealed and heated at 90° C. for 24 h. The ammonia was allowed to evaporate and the residue concentrated to a white solid which was used in the next step without further purification.

Step C: 4-Amino-7-(2-C-hydroxymethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

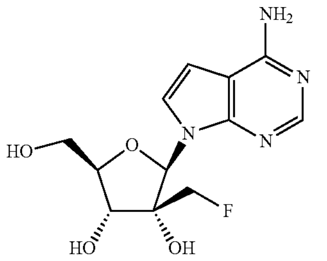
The compound from Step B (120 mg, 0.2 mmol) was dissolved in 1:1 methanol/dichloromethane, 10% Pd—C was added, and the suspension stirred under an H_2 atmosphere for 12 h. The catalyst was removed by filtration through a celite pad and washed with copious amounts of methanol. The combined filtrate was evaporated in vacuo and the residue was purified by flash chromatography (SiO_2, 10% methanol in EtOAc containing 0.1% triethylamine) to give the title compound (50 mg) as a white powder.

^1H NMR (CD_3OD): δ 3.12 (d, 1H, CH_2'), 3.33 (d, 1H, CH_2"), 3.82 (m, 1H, H-5'), 3.99-4.1 (m, 2H, H-4', H-5"), 4.3 (d, 1H, H-3'), 6.2 (s, 1H, H-1'), 6.58 (d, 1H, H-5), 7.45 (d, 1H, H-6), 8.05 (s, 1H, H-2).

LC-MS: Found: 297.2 ($M+H^+$); calc. for $C_{12}H_{16}N_4O_5+H^+$: 297.3.

EXAMPLE 139

4-Amino-7-(2-C-fluoromethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



Step A: 4-Chloro-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C-fluoromethyl-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine

To a solution of the compound from Example 138, Step A (63 mg, 0.1 mmol) in anhydrous dichloromethane (5 mL) under argon, were added 4-dimethylaminopyridine (DMAP) (2 mg, 0.015 mmol) and triethylamine (62 μL, 0.45 mmol). The solution was cooled in an ice bath and p-toluenesulfonyl chloride (30 mg, 0.15 mmol) was added. The reaction was stirred at room temperature overnight, washed with $NaHCO_3$ (2×10 mL), water (10 mL), brine (10 mL), dried over Na_2SO_4 and concentrated to a pink solid in vacuo. The solid was dissolved in anhydrous THF (5 mL) and cooled in an icebath. Tetrabutylammonium fluoride (1M solution in THF, 1 mL, 1 mmol) was added and the mixture stirred at room temperature for 4 h. The solvent was removed in vacuo, the residue taken up in dichloromethane, and washed with $NaHCO_3$ (2×10 mL), water (10 mL) and brine (10 mL). The dichloromethane layer was dried over anhydrous Na_2SO_4, concentrated in vacuo, and purified by flash chromatography (SiO_2, 2:1 hexanes/EtOAc) to afford the title compound (20 mg) as a white solid.

Step B: 4-Amino-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C-fluoromethyl-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine

The compound from Step A (18 mg, 0.03 mmol) was dissolved in the minimum amount of 1,4-dioxane and placed in a stainless steel bomb. The bomb was cooled to −78° C. and liquid ammonia was added. The bomb was sealed and heated at 90° C. for 24 h. The ammonia was allowed to evaporate and the residue concentrated to a white solid which was used in the next step without further purification.

Step C: 4-Amino-7-(2-C-fluoromethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

The compound from Step B (16 mg) was dissolved in 1:1 methanol/dichloromethane, 10% Pd—C was added, and the suspension stirred under an H_2 atmosphere for 12 h. The catalyst was removed by filtration through a celite pad and washed with copious amounts of methanol. The combined filtrate was evaporated in vacuo and the residue was purified by flash chromatography (SiO_2, 10% methanol in EtOAc containing 0.1% triethylamine) to give the title compound (8 mg) as a white powder.

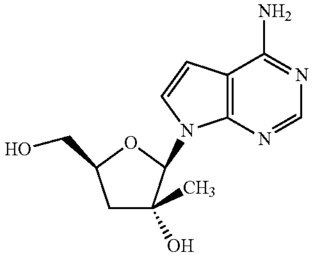
^1H NMR (DMSO-d_6): δ 3.6-3.7 (m, 1H, H-5'), 3.8-4.3 (m, 5H, H-5", H-4', H-3', CH_2) 5.12 (t, 1H, 5'-OH), 5.35 (d, 1H, 3'-OH), 5.48 (s, 1H, 2'-OH), 6.21 (s, 1H, H-1'), 6.52 (d, 1H, H-5), 6.98 (br s, 2H, NH_2), 7.44 (d, 1H, H-6), 8.02 (s, 1H, H-2).

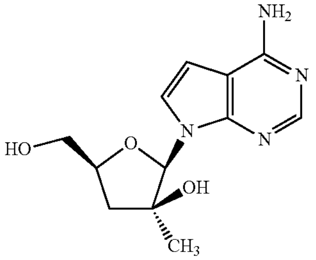
^{19}F NMR (DMSO-d_6): δ −230.2 (t).

ES-MS: Found: 299.1 ($M+H^+$), calc. for $C_{12}H_{15}FN_4O_4+H^+$: 299.27.

EXAMPLES 140 and 141

4-Amino-7-(3-deoxy-2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine and 4-amino-7-(3-deoxy-2-C'-methyl-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]-pyrimidine





Step A: 7-[2,5-Bis-O-(tert-butyldimethylsilyl)-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine and 7-[3,5-Bis-O-(tert-butyldimethylsilyl)-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine

To a stirred solution of tubercidin (5.0 g, 18.7 mmol) in a mixture of pyridine (7.5 mL) and DMF (18.5 mL) was added silver nitrate (6.36 g, 38.8 mmol). This mixture was stirred at room temperature for 2 h. It was cooled in an ice bath and THF (37.4 mL) and tert-butyldimethylsilyl chloride (5.6 g, 37 mmol) was added and the mixture was stirred at room temperature for 2 h. The mixture was then filtered through a pad of celite and washed with THF. The filtrate and washings were diluted with ether containing a small amount of chloroform. The organic layer was washed successively with sodium bicarbonate and water (3×50 mL), dried over anhydrous sodium sulfate and concentrated. The pyridine was removed by coevaporation with toluene and the residue was purified by flash chromatography on silica gel using 5-7% MeOH in CH_2Cl_2 as the eluent; yield 3.0 g.

Step B: 7-[2,5-Bis-O-(tert-butyldimethylsilyl)-β-D-ribofuranosyl)]-4-[di-(4-methoxyphenyl)phenylmethyl]amino-7H-pyrrolo[2,3-d]pyrimidine and 7-[3,5-bis-O-(tert-butyldimethylsilyl)-β-D-ribofuranosyl]-4-[di-(4-methoxyphenyl)phenylmethyl]amino-7H-pyrrolo[2,3-d]pyrimidine

To a solution of mixture of the compounds from Step A (3.0 g, 6.0 mmol) in anhydrous pyridine (30 mL) was added 4,4'-dimethoxytrityl chloride (2.8 g, 8.2 mmol) and the reaction mixture was stirred at room temperature overnight. The mixture was then triturated with aqueous pyridine and extracted with ether. The organic layer was washed with water, dried over anhydrous sodium sulfate and concentrated to a yellow foam (5.6 g). The residue was purified by flash chromatography over silica gel using 20-25% EtOAc in hexanes as the eluent. The appropriate fractions were collected and concentrated to furnish 2',5'-O-bis-O-(tert-butyldimethylsilyl)- and 3',5' bis-O-(tert-butyldimethylsilyl) protected nucleosides as colorless foams (2.2 g and 1.0 g, respectively).

Step C: 7-[2,5-Bis-O-(tert-butyldimethylsilyl)-3-O-tosyl-β-D-ribofuranosyl)]-4-[di-(4-methoxyphenyl)phenylmethyl]amino-7H-pyrrolo[2,3-d]pyrimidine

To an ice-cooled solution of 2',5'-bis-O-(tert-butyldimethylsilyl)-protected nucleoside from Step B (2.0 g, 2.5 mmol) in pyridine (22 mL) was added p-toluenesulfonyl chloride (1.9 g, 9.8 mmol). The reaction mixture was stirred at room temperature for four days. It was then triturated with aqueous pyridine (50%, 10 mL) and extracted with ether (3×50 mL) containing a small amount of CH_2Cl_2 (10 mL). The organic layer was washed with sodium bicarbonate and water (3×30 mL). The organic layer was dried over anhydrous Na_2SO_4 and concentrated. Pyridine was removed by co-evaporation with toluene (3×25 mL). The residual oil was filtered through a pad of silica gel using hexane:ethyl acetate (70:30) as eluent; yield 1.4 g.

Step D: 4-[di-(4-methoxyphenyl)phenylmethyl]amino-7-[3-O-tosyl-β-D-ribofuranosyl-7H-pyrrolo[2,3-d]pyrimidine

A solution of the compound from Step C (1.0 g, 1.1 mmol) and THF (10 mL) was stirred with tetrabutylammonium fluoride (1M solution in THF, 2.5 mL) for 0.5 h. The mixture was cooled and diluted with ether (50 mL). The solution was washed with water (3×50 mL), dried over anhydrous Na_2SO_4, and concentrated to an oil. The residue was purified by passing through a pad of silica gel using hexane:ethyl acetate (1:1) as eluent; yield 780 mg.

Step E: 4-Amino-7-(3-deoxy-2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]-pyrimidine and 4-amino-7-(3-deoxy-2-C-methyl-β-D-arabinofuranosyl)-7H-pyrrolo-[2,3-d]pyrimidine

A solution of CH_3MgI (3.0 M solution in ether, 3.0 mL) in anhydrous toluene (3.75 mL) was cooled in an ice bath. To this was added a solution of the compound from Step D (500 mg, 0.8 mmol) in anhydrous toluene (3.7 mL). The resulting mixture was stirred at room temperature for 3.5 h. It was cooled and treated with aqueous NH_4Cl solution and extracted with ether (50 mL containing 10 mL of CH_2Cl_2). The organic layer was separated and washed with brine (2×30 mL) and water (2×25 mL), dried over anhydrous Na_2SO_4 and concentrated to an oil which was purified by flash chromatography on silica gel using 4% MeOH in CH_2Cl_2 to furnish the 2-C-α-methyl compound (149 mg) and the 2-C-β-methyl compound (34 mg). These derivatives were separately treated with 80% acetic acid and the reaction mixture stirred at room temperature for 2.5 h. The acetic acid was removed by repeated co-evaporation with ethanol and toluene. The residue was partitioned between chloroform and water. The aqueous layer was washed with chloroform and concentrated. The evaporated residue was purified on silica gel using 5-10% MeOH in CH_2Cl_2 as the eluent to furnish the desired compounds as white solids.

4-Amino-7-(3-deoxy-2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine (9.0 mg)

^1H NMR (DMSO-d_6): δ 0.74 (s, 3H, CH_3), 1.77 (dd, 1H, H-3'), 2.08 (t, 1H, H-3"), 3.59 (m, 1H, H-5'), 3.73 (m, 1H, H-5"), 4.15 (m, 1H, H-4'), 5.02 (t, 1H, OH-5'), 5.33 (s, 1H, OH-2'), 6.00 (s, 1H, H-1'), 6.54 (d, 1H, H-7), 6.95 (br s, 2H, NH_2), 7.47 (d, 1H, H-8), 8.00 (s, 1H, H-2); ES-MS: 263.1 [M−H].

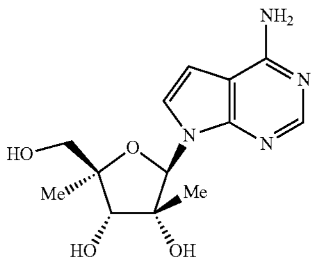
4-Amino-7-(3-deoxy-2-C-methyl-β-D-arabinofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine (15 mg)

^1H NMR (DMSO-d_6): δ 1.23 (s, 3H, CH_3), 2.08 (ddd, 2H, H-3' and **3**"), 3.57 (m, 2H, H-5' and 5"), 4.06 (m, 1H, H-4), 5.10 (s, 1H, OH-2'), 5.24 (t, 1H, OH-5'), 6.01 (s, 1H, H-1'), 6.49 (d, 1H, H-7), 6.89 (br s, 2H, NH_2), 7.35 (d, 1H, H-8), 8.01 (s, 1H, H-2).

ES-MS: 265.2[M+H].

EXAMPLE 142

4-Amino-7-(2,4-C-dimethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



Step A: 5-Deoxy-1,2-O-isopropylidene-D-xylofuranose

1,2-O-Isopropylidene-D-xylofuranose (38.4 g, 0.2 mol), 4-dimethylaminopyridine (5 g), triethylamine (55.7 mL, 0.4 mol) were dissolved in dichloromethane (300 mL). p-Toluenesulfonyl chloride (38.13 g, 0.2 mol) was added and the reaction mixture was stirred at room temperature for 2 h. The reaction mixture was then poured into saturated aqueous sodium bicarbonate (500 mL) and the two layers were separated. The organic layer was washed with aqueous citric acid solution (20%, 200 mL), dried (Na_2SO_4) and evaporated to give a solid (70.0 g). The solid was dissolved in dry THF (300 mL) and $LiAlH_4$ (16.0 g, 0.42 mol) was added in portions over 30 min. The mixture was stirred at room temperature for 15 h. Ethyl acetate (100 mL) was added dropwise over 30 min and the mixture was filtered through a silica gel bed. The filtrate was concentrated and the resulting oil was chromatographed on silica gel (EtOAc/hexane 1/4) to afford the product as a solid (32.5 g).

Step B: 3,5-Bis-O-(2,4-dichlorophenylmethyl)-1-O-methyl-4-methyl-α-D-ribofuranose

Chromium oxide (50 g, 0.5 mol), acetic anhydride (50 mL, 0.53 mol) and pyridine (100 mL, 1.24 mol) were added to dichloromethane (1 L) in an ice water bath and the mixture was stirred for 15 min. 5-Deoxy-1,2-O-isopropylidene-D-xylofuranose (32 g, 0.18 mol) in dichloromethane (200 mL) was added, and the mixture was stirred at the same temperature for 30 min. The reaction solution was diluted with ethyl acetate (1 L) and filtered through a silica gel bed. The filtrate was concentrated to give a yellow oil. The oil was dissolved in 1,4-dioxane (1 L) and formaldehyde (37%, 200 mL). The solution was cooled to 0° C. and solid KOH (50 g) was added. The mixture was stirred at room temperature overnight and was then extracted with ethyl acetate (6×200 mL). After concentration, the residue was chromatographed on silica gel (EtOAc) to afford the product as an oil (1.5 g). The oil was dissolved in 1-methyl-2-pyrrolidinone (20 mL) and 2,4-dichlorophenylmethyl chloride (4 g, 20.5 mmol) and NaH (60%, 0.8 g) were added. The mixture was stirred overnight and diluted with toluene (100 mL). The mixture was then washed with saturated aqueous sodium bicarbonate (3×50 mL), dried ($Na—_2SO_4$) and evaporated. The residue was dissolved in methanol (50 mL) and HCl in dioxane (4 M, 2 mL) was added. The solution was stirred overnight and evaporated. The residue was chromatographed on silica gel (EtOAc/hexane:1/4) to afford the desired product as an oil (2.01 g).

Step C: 3,5-Bis-O-(2,4-dichlorophenylmethyl)-2,4-di-C-methyl-1-O-methyl-α-D-ribofuranose

The product (2.0 g, 4.0 mmol) from Step B and Dess-Martin periodinane (2.0 g) in dichloromethane (30 mL) were stirred overnight at room temperature and was then concentrated under reduced pressure. The residue was triturated with ether ether (50 mL) and filtered. The filtrate was washed with a solution of $Na_2S_2O_3.5H_2O$ (2.5 g) in saturated aqueous sodium bicarbonate solution (50 mL), dried ($MgSO_4$), filtered and evaporated. The residue was dissolved in anhydrous Et_2O (20 mL) and was added dropwise to a solution of MeMgBr in Et_2O (3 M, 10 mL) at −78° C. The reaction mixture was allowed to warm to −30° C. and stirred at −30° C. to −15° C. for 5 h, then poured into saturated aqueous ammonium chloride (50 mL). The two layers were separated and the organic layer was dried ($MgSO_4$), filtered and concentrated. The residue was chromatographed on silica gel (EtOAc/hexane:1/9) to afford the title compound as a syrup (1.40 g).

Step D: 4-Chloro-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2,4-di-C-methyl-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine

To the compound from Step C (0.70 g, 1.3 mmol) was added HBr (5.7 M in acetic acid, 2 mL). The resulting solution was stirred at room temperature for 1 h, evaporated in vacuo and co-evaporated with anhydrous toluene (3×10 mL). 4-Chloro-1H-pyrrolo[2,3-d]pyrimidine (0.5 g, 3.3 mmol) and powdered KOH (85%, 150 mg, 2.3 mmol) were stirred in 1-methyl-2-pyrrolidinone (5 mL) for 30 min and the mixture was co-evaporated with toluene (10 mL). The resulting solution was poured into the above bromo sugar residue and the mixture was stirred overnight. The mixture was diluted with toluene (50 mL), washed with water (3×50 mL) and concentrated under reduced pressure. The residue was chromatographed on silica gel eluting with (EtOAc/Hexane 15/85) to afford a solid (270 mg).

Step E: 4-Amino-7-(2,4-di-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

The compound from Step D (270 mg) was dissolved in dioxane (2 mL) and liquid ammonia (20 g) was added in a stainless steel autoclave. The mixture was heated at 100° C. for 15 h, then cooled and evaporated. The residue was chromatographed on silica gel (EtOAc) to afford a solid (200 mg). The solid (150 mg) and Pd/C (10% 150 mg) in methanol (20 mL) were shaken under H_2 (30 psi) for 3 h, filtered and evaporated. The residue was chromatographed on silica gel ($MeOH/CH_2Cl_2$: 1/9) to afford the desired product as a solid (35 mg).

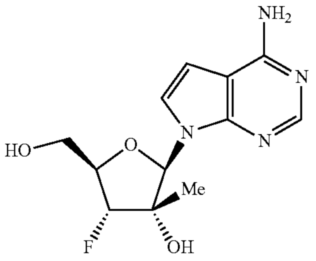
^1H NMR (DMSO-d_6): δ 0.65 (s, 3H), 1.18 (s, 3H), 3.43 (m, 2H), 4.06 (d, 1H, J=6.3 Hz), 4.87 (s, 1H), 5.26 (br, 1H), 5.08 (d, 1H, J=6.3 Hz), 5.25 (t, 1H, J=3.0 Hz), 6.17 (s, 1H), 6.54 (d, 1H, J=3.5 Hz), 6.97 (s, br, 2H), 7.54 (d, 1H, J=3.4 Hz), 8.02 (s, 1H).

^{13}C NMR (DMSO-d_6): δ 18.19, 21.32, 65.38, 73.00, 79.33, 84.80, 90.66, 99.09, 102.41, 121.90, 149.58, 151.48, 157.38.

LC-MS: Found: 295.1 ($M+H^+$); calculated for $C_{13}H_{18}N_4O_4+H^+$: 295.1

EXAMPLE 143

4-Amino-7-(3-deoxy-3-fluoro-2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



Step A: 3-Deoxy-3-fluoro-1-O-methyl-5-O-toluoyl-α-D-ribofuranose

1,2-O-Isopropylidene-D-xylofuranose (9.0 g, 50 mmol) and p-toluoyl chloride (7.0 mL, 50 mmol) in pyridine (50 mL) were stirred for 30 min. Water (10 mL) was added and the mixture was concentrated under reduced pressure. The residue was dissolved in toluene (500 mL) and the solution was washed with water (200 mL) and saturated aqueous sodium bicarbonate (200 mL). The two layers were separated and the organic layer was evaporated. The residue was dissolved in methanol (100 mL) and HCl in dioxane (4 M, 10 mL) was added. The mixture was stirred at room temperature overnight and was then evaporated under reduced pressure. The resulting oil was chromatographed on silica gel (EtOAc/hexane:1/1) to afford an oil (10.1 g). The oil was dissolved in dichloromethane (100 mL) and diethylaminosulfur trifluoride (DAST) (5.7 mL) was added. The mixture was stirred overnight and was then poured into saturated aqueous sodium bicarbonate solution (100 mL). The mixture was extracted with toluene (2×50 mL) and the combined organic layers were concentrated. The residue was chromatographed on silica gel (EtOAc/hexane:15/85) to afford the title compound as an oil (1.50 g).

Step B: 3-Deoxy-3-fluoro-2-C-methyl-1-O-methyl-5-O-toluoyl-α-D-ribofuranose

The product from Step A (1.0 g, 3.5 mmol) and Dess-Martin periodinane (2.5 g) in dichloromethane (20 mL) were stirred overnight at room temperature and was then concentrated under reduced pressure. The residue was triturated with diethyl ether (50 mL) and filtered. The filtrate was washed with a solution of $Na_2S_2O_3.5H_2O$ (12.5 g) in saturated aqueous sodium bicarbonate (100 mL), dried ($MgSO_4$), filtered and evaporated. The residue was dissolved in anhydrous THF (50 mL). $TiCl_a$ (3 mL) and methyl magnesium bromide in ethyl ether (3 M, 10 mL) were added at −78° C. and the mixture was stirred at −50 to −30° C. for 2 h. The mixture was poured into saturated aqueous sodium bicarbonate solution (100 mL) and filtered through Celite. The filtrate was extracted with toluene (100 mL) and evaporated. The residue was chromatographed on silica gel (EtOAc/hexane:15/85) to afford the title compound as an oil (150 mg).

Step C: 4-Amino-7-(3-deoxy-3-fluoro-2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

The product from Step B (150 mg, 0.5 mmol) was dissolved in HBr (30%) in acetic acid (2 mL). After one hour, the mixture was evaporated under reduced pressure and co-evaporated with toluene (10 mL). 4-Chloro-1H-pyrrolo[2,3-d]pyrimidine (0.5 g, 3.3 mmol) and powdered KOH (85%, 150 mg, 2.3 mmol) were stirred in DMF (3 mL) for 30 min and the mixture was co-evaporated with toluene (2 mL). The resulting solution was poured into the above bromo sugar and the mixture was stirred overnight. The mixture was diluted with toluene (50 mL), washed with water (3×50 mL) and concentrated under reduced pressure. The residue was chromatographed on silica gel (EtOAc/hexane 15/85) to afford an oil (60 mg). The oil was dissolved in dioxane (2 mL) and liquid ammonia (20 g) was added in a stainless steel autoclave. The mixture was heated at 85° C. for 18 h, then cooled and evaporated. The residue was chromatographed on silica gel (methanol/dichloromethane:1/9) to afford the title compound as a solid (29 mg).

^1H NMR (DMSO-d_6): δ 0.81 (s, 3H), 3.75 (m, 2H), 4.16 (m, 1H), 5.09 (dd, 1H, J 53.2, 7.8 Hz), 5.26 (br, 1H), 5.77 (s, 1H), 6.15 (d, 1H, J=2.9 Hz), 6.59 (d, 1H, J=3.4 Hz), 7.02 (s br, 2H), 7.39 (d, 1H, J 3.4 Hz), 8.06 (s, 1H).

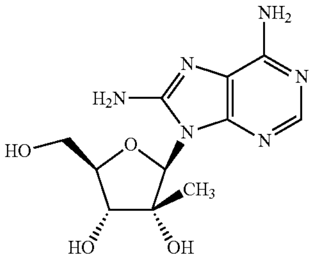
^{13}C NMR (DMSO-d_6): 19.40, 59.56, 77.24, 79.29, 90.15, 91.92, 99.88, 102.39, 121.17, 149.80, 151.77, 157.47.

^{19}F NMR (DMSO-d_6): δ 14.66 (m).

ES-MS: Found: 283.1 ($M+H^+$); calculated for $C_{12}H_{15}FN_4O_3+H^+$: 283.1.

EXAMPLE 144

8-Amino-2'-C-methyladenosine



Step A: 8-Bromo-2'-C-methyladenosine

To a solution of 2'-C-methyladenosine [for preparation, see *J. Med. Chem.* 41: 1708 (1998)] (138 mg, 0.5 mmol) in DMF (4 mL) was added N-bromosuccinimide (231 mg, 1.35 mmol). The solution was stirred protected from light at rt for 2 d and then evaporated in vacuo. The crude product was purified on a silica gel column (3×9 cm) using dichloromethane/methanol (25/1, 20/1 and 15/1) as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (38 mg) as a white solid.

Step B: 8-Amino-2'-C-methyladenosine

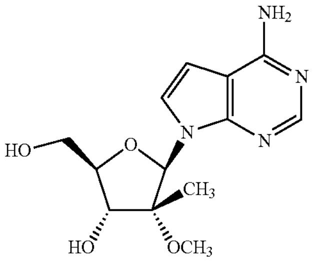
A solution of the compound from Step A (38 mg, 0.11 mmol) in liquid ammonia (10 mL) was heated in a stainless steel autoclave at 105° C. for 1d, then cooled and evaporated. The residue was purified by HPLC [C-18 Phenomenex Luna (10μ; 250×21.2 mm) RP-column; solvents: (A) water, (B) acetonitrile; Linear gradient: 2-35% B in 76 min.] to yield the title compound (12 mg) as a white fluffy material after freeze-drying.

^1H NMR (DMSO-d_6): δ 0.70 (s, 3H, Me), 3.55-3.75 (m, 3H, H-5', H-5'', H-4'), 4.03 (m, 1H, H-3'), 4.81 (s, 1H, 2'-OH), 5.10 (d, 1H, 3'-OH), 5.45 (t, 1H, 5'-OH), 5.86 (s, 1H, H-1'), 6.30, 6.39 (2s, 6H, 2 NH_2), 7.78 (s, 1H, H-2).

ES-MS: Found: 295.0 ($M-H^+$).

EXAMPLE 145

4-Amino-7-(2-C,2-O-dimethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



Step A: 4-chloro-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C,2-O-dimethyl-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine

To a pre-cooled (0° C.) solution of the compound from Example 62, Step D (618 mg, 1.0 mmol) in THF (8 mL) was added methyl iodide (709 mg, 5.0 mmol) and NaH (60% in mineral oil) (44 mg, 1.1 mmol). The resulting mixture was stirred overnight at rt and then poured into a stirred mixture of saturated aqueous ammonium chloride (50 mL) and dichloromethane (50 mL). The organic layer was washed with water (50 mL), dried ($MgSO_4$) and evaporated in vacuo. The resulting crude product was purified on silica gel using ethyl acetate/hexane as the eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (735 mg) as a colorless foam.

Step B: 4-amino-7-[3,5-bis-O-(2,4-dichlorophenylmethyl)-2-C,2-O-dimethyl-β-D-ribofuranosyl]-7H-pyrrolo[2,3-d]pyrimidine

To the compound from Step A (735 mg, 1.16 mmol) was added methanolic ammonia (saturated at 0° C.) (20 mL). The mixture was heated in a stainless steel autoclave at 80° C. overnight, then cooled and the content evaporated in vacuo. The crude mixture was purified on silica gel using ethyl acetate/hexane as the eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (504 mg) as colorless foam.

Step C: 4-amino-7-(2-C,2-O-dimethyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

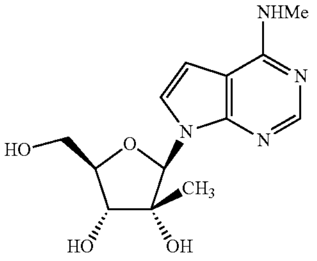
A mixture of the product from Step C (64 mg, 0.1 mmol), MeOH (5 mL), Et_3N (0.2 mL) and 10% Pd/C (61 mg) was hydrogenated on a Parr hydrogenator at 50 psi at room temperature overnight. The mixture was filtered through celite, evaporated in vacuo and filtered through a pad of silica gel using 2% methanol in dichloromethane as eluent. The desired product was collected and evaporated in vacuo. The compound was redissolved in methanol (10 mL) and 10% Pd/C (61 mg) was added. The mixture was hydrogenated on a Parr hydrogenator at 55 psi at room temperature for two weeks. The mixture was filtered through celite, evaporated in vacuo and purified on silica gel using 10% methanol in dichloromethane as eluent. Fractions containing the product were pooled and evaporated in vacuo to give the desired product (110 mg) as a colorless foam.

^1H NMR (DMSO-d_6): δ 0.68 (s, 3H), 3.40 (s, 3H), 3.52-3.99 (overlapping m, 4H), 4.92 (d, 1H), 5.07 (t, 1H), 6.26 (s, 1H), 6.55 (d, 1H), 7.00s br, 2H), 7.46 (d, 1H), 8.05 (s, 1H).

LC-MS: Found: 293.1 (M−H^+); calc. for $C_{12}H_{16}N_4O_4$—H^+: 293.12.

EXAMPLE 146

4-Methylamino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidne



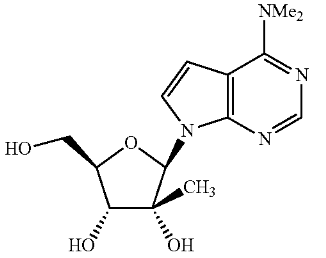
The compound from Step E of Example 62 (200 mg, 0.67 mmol) was added to methylamine (5 mL condensed in a small stainless steel autoclave) and warmed at 85° C. for 48 h, then cooled and evaporated in vacuo. The crude mixture was purified on a silica gel with ethanol as the eluent to give the title compound which separated as an amorphous solid after treatment with MeCN. The amorphous solid was dissolved in water and lyophilized to give a colorless powder (144 mg).

^1H NMR (DMSO-d_6): δ 0.63 (s, 3H, CH_3), 3.32 (s, 3H, NCH_3), 3.58-3.67 (m, 1H, H-5'), 3.79-3.39 (m, 3H, H-5", H-4', H-3'), 5.03 (s, 1H, 2'-OH), 5.04-5.11 (1H, 3'-OH, 1H, 5'-OH), 6.14 (s, 1H, H-1'), 6.58 (d, 1H, $J_{5,6}$=3.6 Hz, H-5), 7.46 (d, 1H, H-6), 7.70 (br s, 1H, NH), 8.14 (s, 1H, H-2).

LC-MS: Found: 295.1 (M−H^+); calc. for $C_{13}H_{18}N_4O_4$+H^+: 294.3.

EXAMPLE 147

4-Dimethylamino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



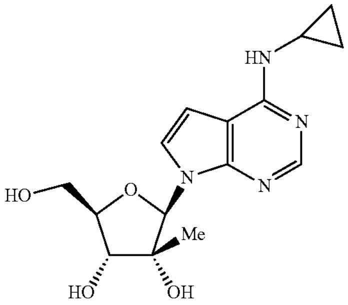
The compound from Step E of Example 62 (200 mg, 0.67 mmol) was added to dimethylamine (5 mL condensed in a small stainless steel autoclave) and warmed at 85° C. for 48 h, then cooled and evaporated in vacuo. The crude mixture was purified on a silica gel with ethanol as the eluent to give the title compound which separated as an amorphous solid after treatment with MeCN. The amorphous solid was dissolved in water and lyophilized to give a colorless powder (164 mg).

^1H NMR (DMSO-d_6): δ 0.64 (s, 3H, CH_3), 3.29 (s, 3H, NCH_3), 3.32 (s, 3H, NCH_3), 3.60-3.66 (m, 1H, H-5'), 3.77-3.97 (m, 3H, H-5", H-4', H-3'), 5.04 (s, 1H, 2'-OH), 5.06-5.11 (1H, 3'-OH, 1H, 5'-OH), 6.21 (s, 1H, H-1'), 6.69 (d, 1H, $J_{5,6}$=3.6 Hz, H-5), 7.55 (d, 1H, H-6), 8.13 (s, 1H, H-2).

LC-MS: Found: 309.3 (M−H^+); calc. for $C_{14}H_{20}N_4O_4$+H^+: 308.33.

EXAMPLE 148

4-Cyclopropylamino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



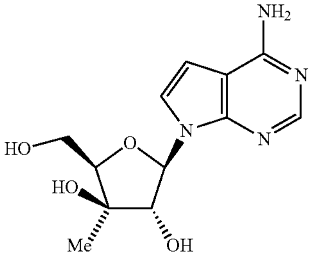
The compound from Step E of Example 62 (200 mg, 0.67 mmol) was added to cyclopropylamine (5 mL condensed in a small stainless steel autoclave) and warmed at 85° C. for 48 h, then cooled and evaporated in vacuo. The crude mixture was purified on a silica gel with ethanol as the eluent to give the title compound which separated as an amorphous solid after treatment with MeCN. The amorphous solid was dissolved in water and lyophilized to give a colorless powder (148 mg).

^{1}H NMR (DMSO-d_6): δ 0.51-0.58 (m, 2H), 0.64 (s, 3H, CH_3), 0.74-0.076 (m, 2H), 3.62-3.67 (m, 1H, H-5'), 3.79-3.82 (m, 3H, H-5"), 3.92-3.96 (m, H-4', H-3'), 5.03 (s, 1H, 2'-OH), 5.05-5.10 (1H, 3'-OH, 1H, 5'-OH), 6.15 (s, 1H, H-1'), 7.48 (d, 1H, $J_{5,6}$=3.6 Hz, H-5), 7.59 (d, 1H, H-6), 8.13 (s, 1H, H-2).

LC-MS: Found: 321.1 (M−H$^+$); calc. for $C_{15}H_{20}N_4O_4+H^+$: 320.3.

EXAMPLE 149

4-Amino-7-(3-C-methyl-β-D-xylofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



Step A: 7-[2,5-Bis-O-(tert-butyldimethylsilyl)-β-D-ribofuranosyl)]-4-[(4-methoxyphenyl)diphenylmethyl]amino-7H-pyrrolo[2,3-d]pyrimidine and 7-[3,5-bis-O-(tert-butyldimethylsilyl)-β-D-ribofuranosyl]-4-[(4-methoxyphenybdiphenylmethyl]amino-7H-pyrrolo[2,3-d]pyrimidine

To a solution of mixture of the compounds from Step A of Examples 140 and 141 (0.32 g, 0.65 mmol) in anhydrous pyridine (6 mL) was added monomethoxytrityl chloride (0.30 g, 0.98 mmol) and the reaction mixture was stirred at room temperature overnight. The mixture was then concentrated and the residue was partitioned between CH_2Cl_2 (70 mL) and water (20 mL). The organic layer was washed with water and brine, dried (Na_2SO_4) and concentrated. The residue was purified on silica gel column using 5-13% EtOAc in hexanes as the eluent. The appropriate fractions were collected and concentrated to furnish 2',5'-bis-O-(tert-butyldimethylsilyl)- and 3',5'-bis-O-(tert-butyldimethylsilyl) protected nucleosides as colorless foams (343 mg and 84 mg, respectively).

Step B: 7-[2,5-Bis-O-(tert-butyldimethylsilyl)-β-D-erythro-pentofuranos-3-ulosyl]-4-[(4-methoxyphenyl)diphenylmethyl]amino-7H-pyrrolo[2,3-d]pyrimidine

To a well-stirred suspension of chromium trioxide (91 mg, 0.91 mmol) in CH_2Cl_2 (4 mL) at 0° C. were added pyridine (147 μL, 1.82 mmol) and then acetic anhydride (86 μL, 0.91 mmol). The mixture was stirred at room temperature for 0.5 h. Then the 2',5'-bis-O-(tert-butyldimethylsilyl) protected nucleoside from step A (343 mg 0.45 mmol) in CH_2Cl_2 (2.5 mL) was added and the mixture stirred at room temperature 2 h. The mixture was then poured into ice-cold EtOAc (10 mL) and filtered through a short silica gel column using EtOAc as the eluent. The filtrate was evaporated and the residue purified on a silica gel column with hexanes and hexanes/EtOAc (7/1) as the eluent to give the title compound (180 mg).

Step C: 7-[2,5-Bis-O-(tert-butyldimethylsilyl)-3-C-methyl-β-D-ribofuranosyl)-4-[(4-methoxyphenyl)diphenylmethyl]amino-7H-pyrrolo[2,3-d]pyrimidine and 7-[2,5-Bis-O-(tert-butyldimethylsilyl)-3-C-methyl-β-D-xylofuranosyl)-4-[(4-methoxyphenyl)diphenylmethyl]amino-7H-pyrrolo[2,3-d]pyrimidine

To a mixture of MeMgBr (3.0 M solution in ether; 0.17 mL, 0.5 mmol) in anhydrous hexanes (1.5 mL) at room temperature was added dropwise a solution of the compound from Step B (78 mg, 0.1 mmol) in anhydrous hexanes (0.5 mL). After 2 h stirring at room temperature, the reaction mixture was poured into ice-cold water (10 mL) and diluted with EtOAc (20 mL), then filtered through Celite which was then thoroughly washed with EtOAc.

The layers were separated and the organic layer was washed with brine, dried (Na_2SO_4) and concentrated. The residue was purified on a silica gel column using 8 to 25% EtOAc in hexanes as eluent to give the 3-C-methyl xylo-(60 mg) and the 3-C-methyl ribo-isomer (20 mg).

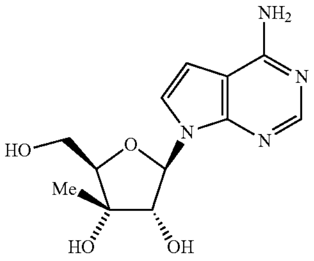
Step D: 4-Amino-7-(3-C-methyl-β-D-xylofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

To an ice-cold solution of 3-C-methyl-xylo isomer from Step C (60 mg, 0.08 mmol) in THF (2 mL) was added TBAF (1 M in THF; 0.32 mL, 0.32 mmol). The reaction mixture was stirred at room temperature for 5 h, then diluted with CH_2Cl_2 (50 mL), washed with water (3×15 mL), dried, and evaporated. The residue was dissolved in dioxane (0.3 mL) and 80% acetic acid (3 mL) was added. The reaction mixture was stirred at room temperature for 1 d and then evaporated. The residue was co-evaporated with dioxane, taken up in water (50 mL) and washed with CH_2Cl_2 (2×10 mL). The aqueous layer was concentrated and then freeze-dried. The residue was purified on silica gel column with CH_2Cl_2/MeOH (20/1 and 10/1) as the eluent to give the title compound as a white fluffy compound after freeze drying (10 mg).

^{1}H NMR (CD_3CN): δ 1.28 (s, 3H, CH_3), 3.56 (br s, 1H, OH), 3.78 (m, 3H, H-4', H-5', H-5"), 4.10 (br s, 1H, OH), 4.44 (d, 1H, J_{21}'=3.9 Hz, H-2'), 5.58 (d, 1H, H-1'), 5.85 (br s, 2H, NH_2), 6.15 (br s, 1H, OH), 6.48 (d, 1H, $J_{5,6}$=3.7 Hz, H-5), 7.23 (d, 1H, H-6), 8.11 (s, 1H, H-2). ES-MS: 281 [MH]$^+$.

EXAMPLE 150

4-Amino-7-(3-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

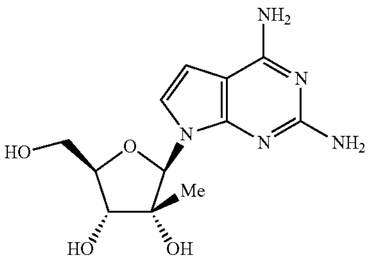


The ribo-isomer (20 mg) from Step C of Example 149 was deprotected using the procedure described in Step D of Example 32 to yield the title compound (4 mg).

^{1}H NMR (CD_3CN): δ 1.43 (s, 3H, CH_3), 3.28 (br s, 1H, OH), 3.58 (m, 2H, H-5', H-5"), 3.99 (m, 1H, H-4'), 4.10 (br s, 1H, OH), 4.62 (d, 1H, $J_{2',1'}$=8.1 Hz, H-2'), 5.69 (d, 1H, H-1'), 5.88 (br s, 3H, OH, NH_2), 6.45 (br s, 1H, OH), 6.51 (d, 1H, $J_{5,6}$=3.7 Hz, H-5), 7.19 (d, 1H, H-6), 8.12 (s, 1H, H-2). ES-MS: 281 $[MH]^+$.

EXAMPLE 151

2,4-Diamino-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



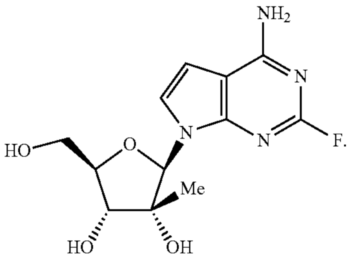
A mixture of the product from Step B of Example 118 (24 mg) in aqueous ammonia (30%, 10 mL) was heated in a stainless steel autoclave at 100° C. overnight, then cooled and evaporated. The residue was purified on a silica gel column with CH_2Cl_2/MeOH (10/1 and 5/1) as the eluent to afford the title compound (15 mg).

^{1}H NMR (DMSO-d_6): δ 0.68 (s, 3H, CH_3), 3.48-3.58 (m 1H, H-5'), 3.68-3.73 (m, 2H, H-5", H-4'), 3.84 (m, 1H, H-3'), 4.72 (s, 1H, 2'-OH), 4.97-5.03 (m, 2H, 3'-OH, 5'-OH), 5.45 (br s, 2H, NH_2), 6.00 (s, 1H, H-1'), 6.28 (d, 1H, J=3.7 Hz, H-5), 6.44 (br s, 2H, NH_2) 6.92 (d, 1H J=3.7 Hz, H-6).

ES MS: 294.1 $(M-H^+)$.

EXAMPLE 152

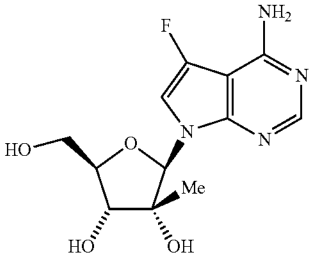
4-Amino-2-fluoro-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



To a solution of HF/pyridine (70%, 2 mL) diluted with pyridine (1 mL) at −30° C. is added the compound of Example 151 (60 mg, 0.2 mmol) in 0.5 mL pyridine followed by tert-butyl nitrite (36 μL, 0.3 mmol). Stirring is continued for 5 min −25° C. Then the solution is poured into ice-water (5 mL), neutralized with 2 N aqueous NaOH, and evaporated to dryness. The residue is purified on a silica gel column with CH_2Cl_2/MeOH (20/1 and 10/1) as the eluent to afford the title compound.

EXAMPLE 153

4-Amino-5-fluoro-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine



Step A: 4-Acetylamino-7-(2,3,5-tri-O-acetyl-2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

To a solution of the compound from step F of Example 62 (280 mg, 1.00 mmol) in pyridine is added acetic anhydride (613 mg, 6.0 mmol). The resulting solution is stirred overnight at ambient temperature evaporated in vacuo and the resulting crude mixture is purified on silica gel using ethyl acetate/hexane as the eluent. Fractions containing the desired product are pooled and evaporated in vacuo to give the desired product.

Step B: 4-Acetylamino-5-bromo-7-(2,3,5-tri-O-acetyl-2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

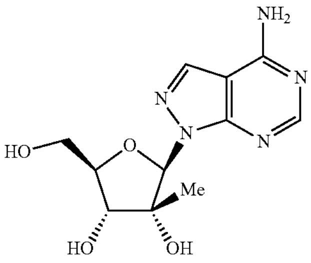
To a pre-cooled (0° C.) solution of the compound from Step A (460 mg, 1.00 mmol) in DMF is added N-bromosuccinimide (178 mg, 1.0 mmol) in DMF. The resulting solution is stirred at 0° C. for 30 min then at room temperature for another 30 min. The reaction is quenched by addition of methanol and evaporated in vacuo. The resulting crude mixture is purified on silica gel using ethyl acetate/hexane as the eluent. Fractions containing the desired product are pooled and evaporated in vacuo to give the desired product.

Step C: 4-Amino-5-fluoro-7-(2-C-methyl-β-D-ribofuranosyl)-7H-pyrrolo[2,3-d]pyrimidine

To a pre-cooled (−78° C.) solution of the compound from Step B (529 mg, 1.00 mmol) in THF is added butyl lithium (2M in hexanes) (0.5 mL, 1.00 mmol). The resulting solution is stirred at −78° C. for 30 min and then quenched with N-fluorobenzensulfonimide (315 mg, 1.00 mmol) in THF. The resulting solution is very slowly allowed to come to ambient temperature and then poured into a stirred mixture of saturated aqueous ammonium chloride and dichloromethane. The organic phase is evaporated in vacuo and treated with ammonium hydroxide at 55° C. in a closed container overnight. The resulting crude mixture is purified on silica gel using dichloromethane/methanol as the eluent. Fractions containing the desired product are pooled and evaporated in vacuo to give the desired product.

EXAMPLE 154

4-Amino-1-(2-C-methyl-β-D-ribofuranosyl)-1H-pyrazolo[3,4-d]pyrimidine



Step A: 4-Amino-1-[3,5-bis-O-(2,4-dichlorophenyl-methyl)-2-C-methyl-β-D-ribofuranosyl]-1H-pyrazolo[3,4-d]pyrimidine

To the compound from Step C of Example 62 (1.00 g, 2.02 mmol) in dichloromethane (20 mL) was bubbled HBr gas for 5 min until it was saturated. The resulting solution was stirred at room temperature for 10 min, evaporated in vacuo and coevaporated with anhydrous toluene (10 mL). 4-Amino-1H-pyrazolo[3,4-d]pyrimidine (Aldrich, 0.43 g, 3.18 mmol) and NaH (60%, 150 mg, 3.8 mmol) were stirred in 1-methyl-2-pyrrolidinone (10 mL) for 30 min. The resulting solution was poured into the above bromo sugar residue and the mixture was stirred overnight. The mixture was diluted with toluene (50 mL), washed with brine (10%, 3×50 mL) and concentrated under reduced pressure. The residue was chromatographed on silica gel (EtOAc as eluent) to afford a solid (400 mg).

Step B: 4-Amino-1-(2-C-methyl-β-D-ribofuranosyl)-1H-pyrazolo[3,4-d]pyrimidine

To a solution of the compound from Step A (0.20 g, 0.33 mmol) in dichloromethane (10 mL) at −78° C. was added boron trichloride (1M in dichloromethane) (3 mL, 3 mmol) dropwise. The mixture was stirred at −78° C. for 0.5 h, then at −45° C. to −30° C. for 2 h. The reaction was quenched by addition of sodium acetate (1.0 g) and methanol (10 mL). The solution was evaporated and the residue was purified by flash chromatography over silica gel using CH_2Cl_2 and CH_2Cl_2/MeOH (95:5-90:10) gradient as the eluent to furnish the desired compound (60 mg) as a solid, which was recrystallized from methanol and acetonitrile to give the title compound as an off-white solid (40 mg).

^{1}H NMR (DMSO-d_6): δ 0.75 (s, 3H), 3.59 (m, 1H), 3.69 (m, 1H), 3.91 (m, 1H), 4.12 (m, 1H), 4.69 (t, 1H, J=5.1 Hz), 5.15 (m, 2H), 6.13 (s, 1H), 7.68 (s, br, 1H), 7.96 (s, br, 1H), 8.18 (s, 1H), 8.21 (s, 1H).

^{13}C NMR (DMSO-d_6): 19.32, 62.78, 74.11, 78.60, 83.65, 90.72, 99.79, 133.50, 153.89, 156.21, 158.05.

LC-MS: Found: 282.1 ($M+H^+$); calculated for $C_{11}H_{15}N_5O_4+H^+$: 282.1.

BIOLOGICAL ASSAYS

The assays employed to measure the inhibition of HCV NS5B polymerase and HCV replication are described below.

The effectiveness of the compounds of the present invention as inhibitors of HCV NS5B RNA-dependent RNA polymerase (RdRp) was measured in the following assay.

A. Assay for Inhibition of HCV NS5B Polymerase:

This assay was used to measure the ability of the nucleoside derivatives of the present invention to inhibit the enzymatic activity of the RNA-dependent RNA polymerase (NS5B) of the hepatitis C virus (HCV) on a heteromeric RNA template.

Procedure:

Assay Buffer Conditions: (50 μL—total/reaction)

- 20 mM Tris, pH 7.5
- 50 μM EDTA
- 5 mM DTT
- 2 mM $MgCl_2$
- 80 mM KCl
- 0.4 U/μL RNAsin (Promega, stock is 40 units/μL)
- 0.75 μg t500 (a 500-nt RNA made using T7 runoff transcription with a sequence from the NS2/3 region of the hepatitis C genome)
- 1.6 μg purified hepatitis C NS5B (form with 21 amino acids C-terminally truncated)
- 1 μM A, C, U, GTP (Nucleoside triphosphate mix)
- [alpha-^{32}P]-GTP or [alpha-^{33}P]-GTP

The compounds were tested at various concentrations up to 100 μM final concentration.

An appropriate volume of reaction buffer was made including enzyme and template t500. Nucleoside derivatives of the present invention were pipetted into the wells of a 96-well plate. A mixture of nucleoside triphosphates (NTP's), including the radiolabeled GTP, was made and pipetted into the wells of a 96-well plate. The reaction was initiated by addition of the enzyme-template reaction solution and allowed to proceed at room temperature for 1-2 h. The reaction was quenched by addition of 20 μL 0.5M EDTA, pH 8.0. Blank reactions in which the quench solution was added to the NTPs prior to the addition of the reaction buffer were included.

50 μL of the quenched reaction were spotted onto DE81 filter disks (Whatman) and allowed to dry for 30 min. The filters were washed with 0.3 M ammonium formate, pH 8 (150 mL/wash until the cpm in 1 mL wash is less than 100, usually 6 washes). The filters were counted in 5-mL scintillation fluid in a scintillation counter.

The percentage of inhibition was calculated according to the following equation:

$$\%\ \text{Inhibition}=[1-(\text{cpm in test reaction}-\text{cpm in blank})/(\text{cpm in control reaction}-\text{cpm in blank})]\times 100.$$

Representative compounds tested in the HCV NS5B polymerase assay exhibited IC_{50}'s less than 100 micromolar.

B. Assay for Inhibition of HCV RNA Replication:

The compounds of the present invention were also evaluated for their ability to affect the replication of Hepatitis C Virus RNA in cultured hepatoma (HuH-7) cells containing a subgenomic HCV Replicon. The details of the assay are described below. This Replicon assay is a modification of that described in V. Lohmann, F. Korner, J-O. Koch, U. Herian, L. Theilmann, and R. Bartenschlager, "Replication of a Subgenomic Hepatitis C Virus RNAs in a Hepatoma Cell Line," *Science* 285:110 (1999).

Protocol:

The assay was an in situ Ribonuclease protection, Scintillation Proximity based-plate assay (SPA). 10,000-40,000 cells were plated in 100-200 μL of media containing 0.8 mg/mL G418 in 96-well cytostar plates (Amersham). Compounds were added to cells at various concentrations up to 100 μM in 1% DMSO at time 0 to 18 h and then cultured for

24-96 h. Cells were fixed (20 min, 10% formalin), permeabilized (20 min, 0.25% Triton X-100/PBS) and hybridized (overnight, 50° C.) with a single-stranded ^{33}P RNA probe complementary to the (+) strand NS5B (or other genes) contained in the RNA viral genome. Cells were washed, treated with RNAse, washed, heated to 65° C. and counted in a Top-Count. Inhibition of replication was read as a decrease in counts per minute (cpm).

Human HuH-7 hepatoma cells, which were selected to contain a subgenomic replicon, carry a cytoplasmic RNA consisting of an HCV 5' non-translated region (NTR), a neomycin selectable marker, an EMCV IRES (internal ribosome entry site), and HCV non-structural proteins NS3 through NS5B, followed by the 3' NTR.

Representative compounds tested in the replication assay exhibited EC$_{50}$'s less than 100 micromolar.

The nucleoside derivatives of the present invention were also evaluated for cellular toxicity and anti-viral specificity in the counterscreens described below.

C. Counterscreens:

The ability of the nucleoside derivatives of the present invention to inhibit human DNA polymerases was measured in the following assays.

a. Inhibition of Human DNA Polymerases Alpha and Beta:

Reaction Conditions:

50 μL reaction volume

Reaction Buffer Components:

20 mM Tris-HCl, pH 7.5
200 μg/mL bovine serum albumin
100 mM KCl
2 mM β-mercaptoethanol
10 mM MgCl$_2$
1.6 μM dA, dG, dC, dTTP
α-^{33}P-dATP

Enzyme and Template:

0.05 mg/mL gapped fish sperm DNA template
0.01 U/μL DNA polymerase α or β

Preparation of Gapped Fish Sperm DNA Template:

Add 5 μL 1M MgCl$_2$ to 500 μL activated fish sperm DNA (USB 70076);
Warm to 37° C. and add 30 μL of 65 U/μL of exonuclease III (GibcoBRL 18013-011);
Incubate 5 min at 37° C.;
Terminate reaction by heating to 65° C. for 10 min;
Load 50-100 μL aliquots onto Bio-spin 6 chromatography columns (Bio-Rad 732-6002) equilibrated with 20 mM Tris-HCl, pH 7.5;
Elute by centrifugation at 1,000×g for 4 min;
Pool eluate and measure absorbance at 260 nm to determine concentration.

The DNA template was diluted into an appropriate volume of 20 mM Tris-HCl, pH 7.5 and the enzyme was diluted into an appropriate volume of 20 mM Tris-HCl, containing 2 mM β-mercaptoethanol, and 100 mM KCl. Template and enzyme were pipetted into microcentrifuge tubes or a 96 well plate. Blank reactions excluding enzyme and control reactions excluding test compound were also prepared using enzyme dilution buffer and test compound solvent, respectively. The reaction was initiated with reaction buffer with components as listed above. The reaction was incubated for 1 hour at 37° C. The reaction was quenched by the addition of 20 μL 0.5M EDTA. 50 μL of the quenched reaction was spotted onto Whatman DE81 filter disks and air dried. The filter disks were repeatedly washed with 150 mL 0.3M ammonium formate, pH 8 until 1 mL of wash is <100 cpm. The disks were washed twice with 150 mL absolute ethanol and once with 150 mL anhydrous ether, dried and counted in 5 mL scintillation fluid.

The percentage of inhibition was calculated according to the following equation:

$$\%\ \text{inhibition}=[1-(\text{cpm in test reaction}-\text{cpm in blank})/(\text{cpm in control reaction}-\text{cpm in blank})]\times 100.$$

b. Inhibition of Human DNA Polymerase Gamma:

The potential for inhibition of human DNA polymerase gamma was measured in reactions that included 0.5 ng/μL enzyme; 10 μM dATP, dGTP, dCTP, and TTP; 2 μCi/reaction [α-^{33}P]-dATP, and 0.4 μg/μL activated fish sperm DNA (purchased from US Biochemical) in a buffer containing 20 mM Tris pH8, 2 mM β-mercaptoethanol, 50 mM KCl, 10 mM MgCl$_2$, and 0.1 μg/μL BSA. Reactions were allowed to proceed for 1 h at 37° C. and were quenched by addition of 0.5 M EDTA to a final concentration of 142 mM. Product formation was quantified by anion exchange filter binding and scintillation counting. Compounds were tested at up to 50 μM.

The percentage of inhibition was calculated according to the following equation:

$$\%\ \text{inhibition}=[1-(\text{cpm in test reaction}-\text{cpm in blank})/(\text{cpm in control reaction}-\text{cpm in blank})]\times 100.$$

The ability of the nucleoside derivatives of the present invention to inhibit HIV infectivity and HIV spread was measured in the following assays.

c. HIV Infectivity Assay

Assays were performed with a variant of HeLa Magi cells expressing both CXCR4 and CCR5 selected for low background β-galactosidase (β-gal) expression. Cells were infected for 48 h, and β-gal production from the integrated HIV-1 LTR promoter was quantified with a chemiluminescent substrate (Galactolight Plus, Tropix, Bedford, Mass.). Inhibitors were titrated (in duplicate) in twofold serial dilutions starting at 100 μM; percent inhibition at each concentration was calculated in relation to the control infection.

d. Inhibition of HIV Spread

The ability of the compounds of the present invention to inhibit the spread of the human immunodeficiency virus (HIV) was measured by the method described in U.S. Pat. No. 5,413,999 (May 9, 1995), and J. P. Vacca, et al., *Proc. Natl. Acad. Sci.*, 91: 4096-4100 (1994), which are incorporated by reference herein in their entirety.

The nucleoside derivatives of the present invention were also screened for cytotoxicity against cultured hepatoma (HuH-7) cells containing a subgenomic HCV Replicon in an MTS cell-based assay as described in the assay below. The HuH-7 cell line is described in H. Nakabayashi, et al., *Cancer Res.*, 42: 3858 (1982).

e. Cytotoxicity Assay:

Cell cultures were prepared in appropriate media at concentrations of approximately 1.5×10^5 cells/mL for suspension cultures in 3 day incubations and 5.0×10^4 cells/mL for adherent cultures in 3 day incubations. 99 μL of cell culture was transferred to wells of a 96-well tissue culture treated plate, and 1 μL of 100-times final concentration of the test compound in DMSO was added. The plates were incubated at 37° C. and 5% CO$_2$ for a specified period of time. After the incubation period, 20 μL of CellTiter 96 Aqueous One Solution Cell Proliferation Assay reagent (MTS) (Promega) was added to each well and the plates were incubated at 37° C. and 5% CO$_2$ for an additional period of time up to 3 h. The plates were agitated to mix well and absorbance at 490 nm was read using a plate reader. A standard curve of suspension culture cells was prepared with known cell numbers just prior to the addition of MTS reagent. Metabolically active cells reduce MTS to formazan. Formazan absorbs at 490 nm. The absorbance at 490 nm in the presence of compound was compared to absorbance in cells without any compound added.

Reference:

Cory, A. H. et al., "Use of an aqueous soluble tetrazolium/formazan assay for cell growth assays in culture," *Cancer Commun.* 3: 207 (1991).

The following assays were employed to measure the activity of the compounds of the present invention against other RNA-dependent RNA viruses:

a. Determination of In Vitro Antiviral Activity of Compounds Against Rhinovirus (Cytopathic Effect Inhibition Assay):

Assay conditions are described in the article by Sidwell and Huffman, "Use of disposable microtissue culture plates for antiviral and interferon induction studies," *Appl. Microbiol.* 22: 797-801 (1971).

Viruses:

Rhinovirus type 2 (RV-2), strain HGP, was used with KB cells and media (0.1% $NaHCO_3$, no antibiotics) as stated in the Sidwell and Huffman reference. The virus, obtained from the ATCC, was from a throat swab of an adult male with a mild acute febrile upper respiratory illness. Rhinovirus type 9 (RV-9), strain 211, and rhinovirus type 14 (RV-14), strain Tow, were also obtained from the American Type Culture Collection (ATCC) in Rockville, Md. RV-9 was from human throat washings and RV-14 was from a throat swab of a young adult with upper respiratory illness. Both of these viruses were used with HeLa Ohio-1 cells (Dr. Fred Hayden, Univ. of VA) which were human cervical epitheloid carcinoma cells. MEM (Eagle's minimum essential medium) with 5% Fetal Bovine serum (FBS) and 0.1% $NaHCO_3$ was used as the growth medium.

Antiviral test medium for all three virus types was MEM with 5% FBS, 0.1% $NaHCO_3$, 50 n gentamicin/mL, and 10 mM $MgCl_2$.

2000 μg/mL was the highest concentration used to assay the compounds of the present invention. Virus was added to the assay plate approximately 5 min after the test compound. Proper controls were also run. Assay plates were incubated with humidified air and 5% CO_2 at 37° C.

Cytotoxicity was monitored in the control cells microscopically for morphologic changes. Regression analysis of the virus CPE data and the toxicity control data gave the ED50 (50% effective dose) and CC50 (50% cytotoxic concentration). The selectivity index (SI) was calculated by the formula: $SI=CC50 \div ED50$.

b. Determination of In Vitro Antiviral Activity of Compounds Against Dengue, Banzi, and Yellow Fever (CPE Inhibition Assay)

Assay details are provided in the Sidwell and Huffman reference above.

Viruses:

Dengue virus type 2, New Guinea strain, was obtained from the Center for Disease Control.

Two lines of African green monkey kidney cells were used to culture the virus (Vero) and to perform antiviral testing (MA-104). Both Yellow fever virus, 17D strain, prepared from infected mouse brain, and Banzi virus, H 336 strain, isolated from the serum of a febrile boy in South Africa, were obtained from ATCC. Vero cells were used with both of these viruses and for assay.

Cells and Media:

MA-104 cells (BioWhittaker, Inc., Walkersville, Md.) and Vero cells (ATCC) were used in Medium 199 with 5% FBS and 0.1% $NaHCO_3$ and without antibiotics.

Assay medium for dengue, yellow fever, and Banzi viruses was MEM, 2% FBS, 0.18% $NaHCO_3$ and 50 μg gentamicin/mL.

Antiviral testing of the compounds of the present invention was performed according to the Sidwell and Huffman reference and similar to the above rhinovirus antiviral testing. Adequate cytopathic effect (CPE) readings were achieved after 5-6 days for each of these viruses.

c. Determination of In Vitro Antiviral Activity of Compounds Against West Nile Virus (CPE Inhibition Assay)

Assay details are provided in the Sidwell and Huffman reference cited above. West Nile virus, New York isolate derived from crow brain, was obtained from the Center for Disease Control.

Vero cells were grown and used as described above. Test medium was MEM, 1% FBS, 0.1% $NaHCO_3$ and 50 μg gentamicin/mL.

Antiviral testing of the compounds of the present invention was performed following the methods of Sidwell and Huffman which are similar to those used to assay for rhinovirus activity. Adequate cytopathic effect (CPE) readings were achieved after 5-6 days.

d. Determination of In Vitro Antiviral Activity of Compounds Against Rhino, Yellow Fever, Dengue, Banzi, and West Nile Viruses (Neutral Red Uptake Assay)

After performing the CPE inhibition assays above, an additional cytopathic detection method was used which is described in "Microtiter Assay for Interferon: Microspectrophotometric Quantitation of Cytopathic Effect," *Appl. Environ. Microbiol.* 31: 35-38 (1976). A Model EL309 microplate reader (Bio-Tek Instruments Inc.) was used to read the assay plate. ED50's and CD50's were calculated as above.

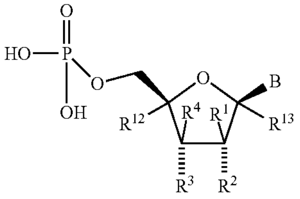
EXAMPLE OF A PHARMACEUTICAL FORMULATION

As a specific embodiment of an oral composition of a compound of the present invention, 50 mg of Example 61 or 62 is formulated with sufficient finely divided lactose to provide a total amount of 580 to 590 mg to fill a size O hard gelatin capsule.

While the invention has been described and illustrated in reference to specific embodiments thereof, those skilled in the art will appreciate that various changes, modifications, and substitutions can be made therein without departing from the spirit and scope of the invention. For example, effective dosages other than the preferred doses as set forth hereinabove may be applicable as a consequence of variations in the responsiveness of the human being treated for severity of the HCV infection. Likewise, the pharmacologic response observed may vary according to and depending upon the particular active compound selected or whether there are present pharmaceutical carriers, as well as the type of formulation and mode of administration employed, and such expected variations or differences in the results are contemplated in accordance with the objects and practices of the present invention. It is intended therefore that the invention be limited only by the scope of the claims which follow and that such claims be interpreted as broadly as is reasonable.

What is claimed is:

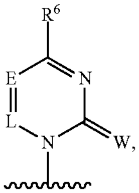
1. A compound having the formula:


(VIII)

or a pharmaceutically acceptable salt thereof,

wherein:

B is:



L is CH or N;

E is N or CR^5;

W is O or S;

R^1 is C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms; R^3 is hydroxy or C_{1-4} alkoxy; and R^2 is selected from the group consisting of

halogen,

C_{1-4} alkyl, optionally substituted with 1 to 3 fluorine atoms,

C_{1-10} alkoxy, optionally substituted with C_{1-3} alkoxy or 1 to 3 fluorine atoms,

C_{2-6} alkenyloxy,

C_{1-4} alkylthio,

C_{1-8} alkylcarbonyloxy,

aryloxycarbonyl,

azido,

amino,

C_{1-4} alkylamino, and

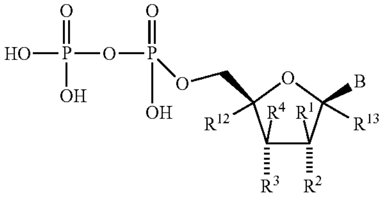
di(C_{1-4} alkyl)amino;

R^4 and R^6 are each independently H, OH, SH, NH_2, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, C_{3-6} cycloalkylamino, halogen, C_{1-4} alkyl, C_{1-4} alkoxy, or CF_3;

R^5 is H, C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-4} alkylamino, CF_3, or halogen;

R^{12} and R^{13} are each independently hydrogen or methyl.

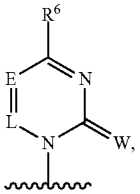
2. A compound having the formula:


(IX)

or a pharmaceutically acceptable salt thereof,

wherein:

B is:



L is CH or N;

E is N or CR^5;

W is O or S;

R^1 is C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms; R^3 is hydroxy or C_{1-4} alkoxy; and R^2 is selected from the group consisting of

halogen,

C_{1-4} alkyl, optionally substituted with 1 to 3 fluorine atoms,

C_{1-10} alkoxy, optionally substituted with C_{1-3} alkoxy or 1 to 3 fluorine atoms,

C_{2-6} alkenyloxy,

C_{1-4} alkylthio,

C_{1-8} alkylcarbonyloxy,

aryloxycarbonyl,

azido,

amino,

C_{1-4} alkylamino, and

di(C_{1-4} alkyl)amino;

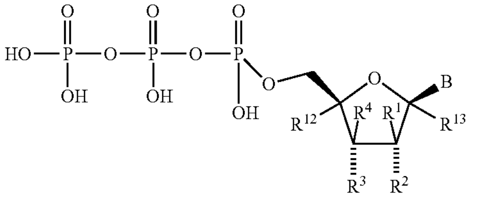
R^4 and R^6 are each independently H, OH, SH, NH_2, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, C_{3-6} cycloalkylamino, halogen, C_{1-4} alkyl, C_{1-4} alkoxy, or CF_3;

R^5 is H, C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-4} alkylamino, CF_3, or halogen;

and

R^{12} and R^{13} are each independently hydrogen or methyl.

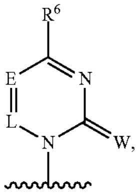
3. A compound having the formula:


(VII)

or a pharmaceutically acceptable salt or thereof,

wherein:

B is:



L is CH or N;

E is N or CR^5;

W is O or S;

R^1 is C_{2-4} alkenyl, C_{2-4} alkynyl, or C_{1-4} alkyl optionally substituted with amino, hydroxy, or 1 to 3 fluorine atoms; R^3 is hydroxy or C_{1-4} alkoxy; and R^2 is selected from the group consisting of

- halogen,
- C_{1-4} alkyl, optionally substituted with 1 to 3 fluorine atoms,
- C_{1-10} alkoxy, optionally substituted with C_{1-3} alkoxy or 1 to 3 fluorine atoms,
- C_{2-6} alkenyloxy,
- C_{1-4} alkylthio,
- C_{1-8} alkylcarbonyloxy,
- aryloxycarbonyl,
- azido,
- amino,
- C_{1-4} alkylamino, and
- di(C_{1-4} alkyl)amino;

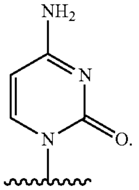
R^4 and R^6 are each independently H, OH, SH, NH_2, C_{1-4} alkylamino, di(C_{1-4} alkyl)amino, C_{3-6} cycloalkylamino, halogen, C_{1-4} alkyl, C_{1-4} alkoxy, or CF_3;

R^5 is H, C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-4} alkylamino, CF_3, or halogen;

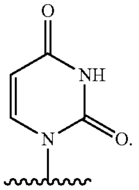
and

R^{12} and R^{13} are each independently hydrogen or methyl.

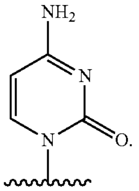
4. The compound of claim **1**, wherein B is:



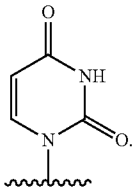
5. The compound of claim **1**, wherein B is:



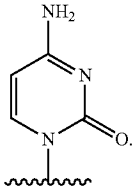
6. The compound of claim **2**, wherein B is:



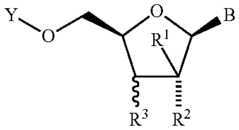
7. The compound of claim **2**, wherein B is:



8. The compound of claim **3**, wherein B is:



9. A compound having the formula:

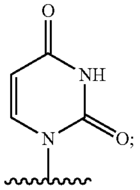


(III)

or a pharmaceutically acceptable salt thereof,

wherein:

B is



Y is $P_3O_9H_4$ or $P_2O_6H_3$;

R^1 is C_{1-4} alkyl;

R^2 is fluoro;

R^3 is OH or C_{1-4} alkoxy.

10. The compound of claim **9**, or a pharmaceutically acceptable salt thereof, wherein Y is $P_3O_9H_4$.

11. The compound of claim **9**, or a pharmaceutically acceptable salt thereof, wherein Y is $P_2O_6H_3$.

* * * * *

CERTIFICATE OF SERVICE

I certify that today, November 29, 2016, I electronically filed the foregoing Corrected Brief for Defendants-Appellants Merck & Co., Inc., Merck Sharp & Dohme Corp., and Isis Pharmaceuticals, Inc. with the Clerk of the Court for the U.S. Court of Appeals for the Federal Circuit using the appellate CM/ECF system. All participants in the case are registered CM/ECF users and will be served by the appellate CM/ECF system.

November 29, 2016 /s/ Jeffrey A. Lamken

CERTIFICATE OF COMPLIANCE

1. This brief complies with the type-volume limitation of Fed. R. App. P. 32(a)(7)(B) because:

_X_ this brief contains 13,991 words, excluding the parts of the brief exempted by Fed. R. App. P. 32(a)(7)(B)(iii), or

___ this brief uses a monospaced typeface and contains [state the number of] lines of text, excluding the parts of the brief exempted by Fed. R. App. P. 32(a)(7)(B)(iii).

2. This brief complies with the typeface requirements of Fed. R. App. P. 32(a)(5) and the type-style requirements of Fed. R. App. P. 32(a)(6) because:

_X_ this brief has been prepared in a proportionally spaced typeface using Microsoft Word in Times New Roman 14-point font, or

___ this brief has been prepared in a monospaced typeface using [state name and version of word processing program] with [state number of characters per inch and name of type style].

November 23, 2016 /s/ Jeffrey A. Lamken